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Organocatalysis and Biocatalysis hand in hand: combining catalysts in one-pot procedures

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Abstract: Nature has developed multi-step processes catalysed by several catalysts working concurrently, thus improving reaction efficiency. The quest for novel and improved catalytic systems has led to the development of biocatalytic and later to organocatalytic procedures as very valuable tools in asymmetric synthesis while using mild reaction conditions in absence of metal catalysts. As a timeless challenge, chemists are facing the need for process designs in which different sort of catalysts can operate successfully in one-pot concurrent fashion. Likewise, such designs bring about the best of each catalyst and, in certain cases, enable to improve problematic issues, such as reactivity, selectivity, solubility, inhibition, *etc*. Specifically, to combine these two types of catalysts in one-pot, achieving high yields and selectivity, is a fascinating edge of catalysis. This review covers representative advances in this field, in particular those in which biocatalysts and organocatalysts are employed either in sequential reactions or in simultaneous processes.

Keywords: One-pot procedures; Multistep synthesis; Organic catalysis; Enzyme catalysis; Enantioselectivity; Concurrent processes.

1.Introduction

In biological systems, nature has developed evolutionary pressure for the optimization of a given process. Resource- and energy-saving maximization became evolutionary pressures; hence, cellular machineries, *i.e.* enzymatic networks dedicated to a specific or general cellular task, have evolved. In order to improve metabolic efficiency, living systems make use of several extremely selective catalysts working at the same time, thus forming complex biochemical networks. To achieve such a degree of success, a perfect regulation of the different catalysts working concomitantly, often in the same compartment, became of utmost importance. In this way, for a given biosynthetic pathway, the product of one reaction is the substrate of the following one, avoiding intermediate accumulation and, therefore, side reactions.[1]

Often, chemists employ consecutive multistep chemical synthesis with catalytically efficient reactions thus improving the overall ‘‘atom economy’’ of the process.[2] These methodologies have been widely adopted in the industrial manufacture of fine chemicals and pharmaceutical intermediates. In this frame, strategies where multiple catalysts simultaneously work in ‘‘one-pot,’’ avoiding the costly isolation and purification of chemical intermediates, are named concurrent.[1a,3] Therefore, reactions taking place in cellular environment are in fact considered as concurrent. In organic chemistry, it has been largely demonstrated that running multiple reactions in one-pot, either in sequential (also known as stepwise, when operations such as addition of catalysts/reagents, temperature/atmosphere modification, *etc*. are made during the course of the process to ensure the proper reactivity mode) and simultaneous (also known as cascade or domino reactions, in which conditions are not modified during the process and catalysts/reagents are added at the beginning, thus requiring only one operational step) mode, results challenging to a great extent given the diverging reaction conditions suitable for each single transformation.[4] Thus, tremendous efforts are made in order to find suitable conditions to harmonically combine multiple catalytic reactions with no cross-spoiling effects.[5]

In a broad sense, organocatalysis can be defined as the acceleration of a chemical reaction using an organic compound in substoichiometric amounts in the absence of (transition) metals.[6] On the other hand, an acceptable definition of biocatalysis in organic synthesis can be the employment of a biomolecule (protein, antibody, ribozyme) or a living organism to carry out the transformation of a (non-)natural organic compound.[7]

Nowadays, given that both organo- and biocatalysis are becoming mature and mechanistic

understanding of enzymatic reactions along with catalytically-productive organic associations have been deeply understood, it is rather remarkable that the number of examples where both sort of catalysis working concurrently were successfully applied is limited when compared with the far more explored combination of metal-catalysed processes and biocatalysis.[8] Some reports have been published based on the combination of transition metals and organocatalysts;[9] but not so many examples are available dealing with combination of organocatalysis and biocatalysis in one-pot procedures.[10]

It must be taken into account that organic substances are not always soluble in aqueous media, so a proper solvent selection is usually an issue in both organo- and biocatalytic systems. Notwithstanding, the use of cosolvents or additives (in the frame of 'medium engineering' concept), is a common practice in those catalytic methodologies, thus circumventing solubility and, in certain cases, reactivity problems.[11] For a combination of organo- and biocatalytic processes in one-pot procedures, the reaction medium must be carefully tailored in order to avoid cross-spoiling. Cosolvents and additives for the organocatalysed reaction, need to be tested towards the biocatalysed reaction to prevent inhibitory effects.

From a synthetic point of view, chirality in the formed product can be installed or defined either through the enzymatic or the organocatalysed reaction, or, even, both catalytic systems may define stereocentres in the same catalytic cycle, increasing the complexity of the final products.

Fabricio R. Bisogno obtained his degree in Biochemistry (2004) at San Luis National University (Argentina). In 2007, he spent half a year at the Weizmann Institute of Sciences (Israel, Prof. Meir Lahav) involved in research on the origin of homochirality. Then, he pursued PhD studies at University of Oviedo (Spain, Prof. Iván Lavandera and Prof. Vicente Gotor), dealing with biocatalytic redox processes (2010). Besides, in 2012 he obtained another PhD degree at San Luis National University (Argentina, Prof. Marcela Kurina-Sanz) working on fungal biotransformation of bioactive compounds. After postdoc periods in Oviedo and Córdoba (Argentina), in 2013 he joined the Prof Alicia Peñéñory's group as researcher at Córdoba National University and INFIQC-CONICET (Argentina). His research interest comprises combination of biocatalysis with metal- and organocatalysis for the construction of cooperative systems, along with exploration of novel reactivities for organo-sulfur and –selenium containing compounds in enzymatic or biomimetic processes.

Martín G. López-Vidal obtained his degree in Biochemistry at Córdoba National University (2014, Argentina). In 2015, he joined the Prof Alicia Peñéñory's group as a PhD CONICET-fellow, under the guidance of Fabricio R. Bisogno at Córdoba National University. In 2016 he spent a three-months stage in the lab of Biocatalysis at the University of Graz (Austria) exploring the reactivity of sulfur-containing compounds with ene-reductases under the supervision of Prof Mélanie Hall. His research interest is focused on development of novel chemoenzymatic cascades for stereoselective preparation of organochalcogen-compounds.

Gonzalo de Gonzalo obtained his Ph.D. in 2003 (Prof. Vicente Gotor, University of Oviedo) working on the field of biocatalysis employing lipases and oxynitrilases. He spent his postdoctoral research at Consiglio Nazionale delle Ricerche (ICRM, Milano, Italy, Dr. Giacomo Carrea), moving back to University of Oviedo with a Juan de la Cierva Fellowship. After a one-year postdoctoral stage at University of Groningen (The Netherlands, Prof. Marco W. Fraaije) working in the research of novel oxidative biocatalysts, he spent two years at the R&D Department of the pharmaceutical company Antibióticos S.A.U. (León, Spain). He is currently a Ramón and Cajal Researcher (MINECO) at University of Sevilla. His research is focused on asymmetric synthesis by using different approaches, including biocatalytic and organocatalytic procedures, as well as the development of concurrent chemo- and biocatalytic reactions.

With the settlement of technologies such as photochemistry, flow chemistry, mechanochemistry, among other, that can be incorporated into complex multicatalytic processes, the boundaries of combined catalysis are continuously pushed forward.

In this review, focus will lie on selected representative examples that may give a general idea of the possibilities of combining organocatalysts and biocatalysts in one-pot including sequential and simultaneous methodologies. Special emphasis will be given to asymmetric processes.

2 Sequential reactions employing organocatalysts and biocatalysts

Initially, we shall deal with processes in which the organocatalyst and the biocatalyst are able to catalyse sequential reactions in order to have multistep one-pot procedures (Figure 1).[12] The selectivity of the final products when chiral compounds are synthesized, can be induced by only the organocatalyst, only the biocatalyst or by both

catalysts performing selective reactions in the sequential process.

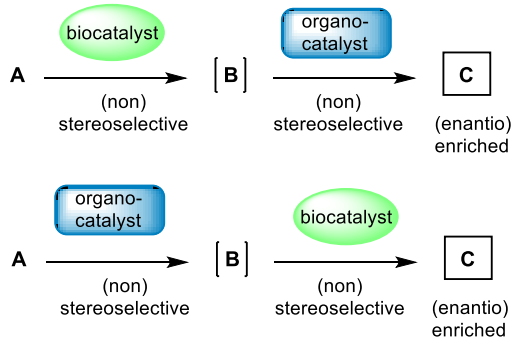
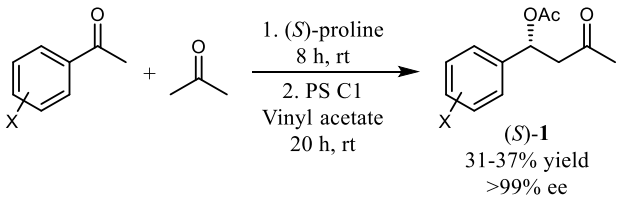


Figure 1. Schematic representation of a sequential one-pot process using organocatalysis and biocatalysis.

One of the first examples described in literature of sequential multistep one-pot process combining organocatalysts and enzymes, was described in 2004 for the synthesis of enantiomerically pure aldols, as shown in Scheme 1.[13]

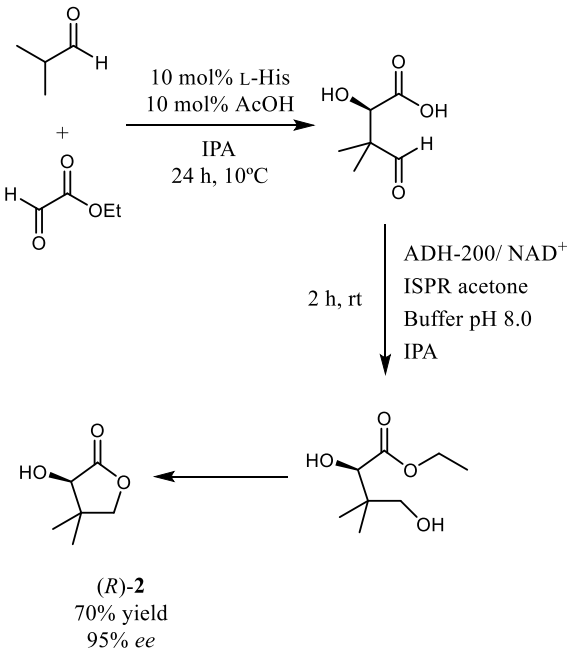
In this process, the proline-catalysed aldol reaction between aromatic aldehydes and acetone was combined with the kinetic resolution of the resulting alcohols catalysed by lipases in presence of vinyl acetate. Aldol reactions were performed in neat acetone, as the usual solvents for organocatalysed-aldol reactions are toxic to lipases. Under these conditions, the aldol adducts were obtained with good yields and moderate selectivities. Such adducts were tested as substrates in biocatalysed acylations using vinyl acetate as acyl donor. Lipase form *Pseudomonas cepacia* Amano I (PS C1) led to excellent selectivities in the kinetic resolutions, thus improving the *ee* obtained in the aldol reaction. In view of these results, the processes were carried out in one-pot. Acetone was removed from the reaction medium before the addition of vinyl acetate and the lipase. Likewise, the use of (*S*)-proline and lipase PS C1 led to the formation of the enantiopure (*S*)-acetates (*S*)-**1** with moderate yields (around 30-40%), These yields achieved in the one-pot procedures were slightly lower than those obtained in the stepwise reactions.



Scheme 1. Synthesis of optically active aldols by combining an organocatalytic aldol reaction with a biocatalysed acylation in a sequential one-pot process.[13]

Other amino acids different from proline and its derivatives have been also studied as catalysts in aldol reactions. Thus, L-histidine-catalysed aldol reaction combined with alcohol dehydrogenase (ADH) catalysed reduction,[14] has been reported towards the asymmetric synthesis of (*R*)-pantolactone (**2**).[15]

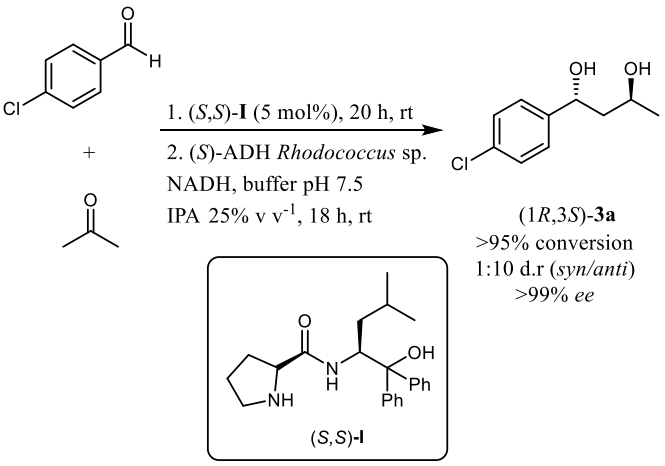
Firstly, organocatalysed aldol reaction between isobutanal and ethyl glyoxylate in 2-propanol (IPA) was explored in order to obtain the enantioenriched aldol-adduct. Several catalysts were studied, and among the tested amino acids and amino(thio)urea catalysts, L-histidine was chosen, displaying good activity with high enantioselectivity. An additional advantage of L-histidine towards more complex organocatalysts is its commercial availability in both enantiomeric forms. In the Brønsted acid cocatalyst screening, turned out that acids with pKa-values between 4.0-5.0 showed best results and the authors adopted acetic acid as cocatalyst. It has been described that those acids could increase the addition yield by facilitating the hydrolysis of the iminium ion formed between substrate and catalyst.[16] Reactions were performed using ethyl glyoxylate 2.5 M, 10 mol% of L-histidine, 10 mol% of acetic acid in a mixture water/alcohol 1:1 for 24 h at 10°C leading to the formation of the (*R*)-α-hydroxyester up to 95% conversion, 85% yield and 79% *ee*. Further studies were focused on the enzymatic reduction of the aldol adduct (0.5 M)and further spontaneous lactonization of the hydroxyester, where ADH-200 from Evocatal (evo-1.1.200) showed higher conversions at pH 8.0 in buffer/2-propanol 20% v v^{-1} up to 67 % conversion and 95% *ee*. Removal of volatile compounds such as acetone (a byproduct formed in the nicotinamide cofactor NAD^+ recycling) and non-converted aldehydes prior to the bioreduction step increases significantly the conversion (64% *vs*. 33% without *in vacuo* removal of volatile compounds), probably due competition of these compounds for aldol adduct. In order to *in situ* remove the acetone formed as side-product of the co-factor regeneration system (*In Situ Product Removal* – ISPR),[17] continuous air saturated with water/2-propanol (5 % v v^{-1}) passed through the bioreduction has proved to be an effective strategy leading to 86 % of conversion of the aldol product and 95 % *ee* after 2 h. The entire chemoenzymatic synthesis of (*R*)-pantolactone was performed in what the authors claimed as a "one-pot-like" process, as the volatile compounds of the aldol reaction are removed *in vacuo* prior to the bioreduction and ISPR during enzymatic reaction. Starting from 1.28 mmol of ethyl glyoxate and 2.65 mmol of isobutanal in 0.41 mL of IPA, (*R*)-**2** was obtained with 55% of conversion (referred to ethylglyoxylate) and 95% *ee*, as shown in Scheme 2.



Scheme 2. One-pot preparation of (*R*)-pantolactone **2** by combining organocatalysed aldol reaction with enzymatic bioreduction in presence of ADH-200.[15]

In 2009, the sequential one-pot synthesis of chiral 1,3-diols combining an organocatalytic aldol reaction and an enzymatic ketoreduction has been carried out in aqueous media (Scheme 3).[18] The proline-based catalyst **I**, developed by Singh in both homochiral diastereomeric forms,[19] has demonstrated to be an efficient catalyst for the enantioselective formation of β-hydroxyketones with high yields and enantiomeric excesses. For this reason, preliminary studies were focused on the optimisation of organocatalysed aldol reaction using both (*S*,*S*)-**I** and (*R*,*R*)-**I** as catalysts. Thus, employing *p*-chlorobenzaldehyde and acetone as model substrates, both enantiomeric forms of the corresponding β-hydroxyketone were achieved with the same enantiomeric excess (83%). However, the obtained yields were lower in case of the (*R*)-isomer than the (*S*)-isomer (58 and 71%, respectively). Further studies were focused on the bioreduction of the aldol products. Either (*S*)-ADH from *Rhodococcus* sp. or (*R*)-ADH from *Lactobacillus kefir* using 2-propanol as cosubstrate in substrate-coupled cofactor recycling system, allowed to synthesise the corresponding isomeric four possible 1,3-diols **3** with excellent results (>95% of conversion, >99% *ee* and 1:11 of diastereomeric ratio).

In order to explore the substrate scope of this process, other substituents at the *para*-position of the aromatic ring were tested, where *p*-tolualdehyde showed similar results with only a slight decrease in the stereoselectivity of the aldol reaction (76% *ee*). Once the individual steps were optimised, for the proof-of-concept of the whole one-pot two-steps system, *p*-chlorobenzaldehyde (0,5 mmol) and acetone were chosen as substrates. Thus, corresponding β-hydroxyketone was attained using (*S*,*S*)-**I** in 20 hours at r.t. After the aldol reaction, bioreduction employing the (*S*)-ADH was carried out in phosphate buffer pH 7.0 (67 mM of the substrate) containing 2-propanol 25% v v^{-1} as cosubstrate at r.t., affording the desired (1*R*,3*S*)-**3a** with >99% *ee*, 1:10 *syn*/*anti* diastereomeric ratio after 18 hours, reaching >95% of conversion as shown in Scheme 3.



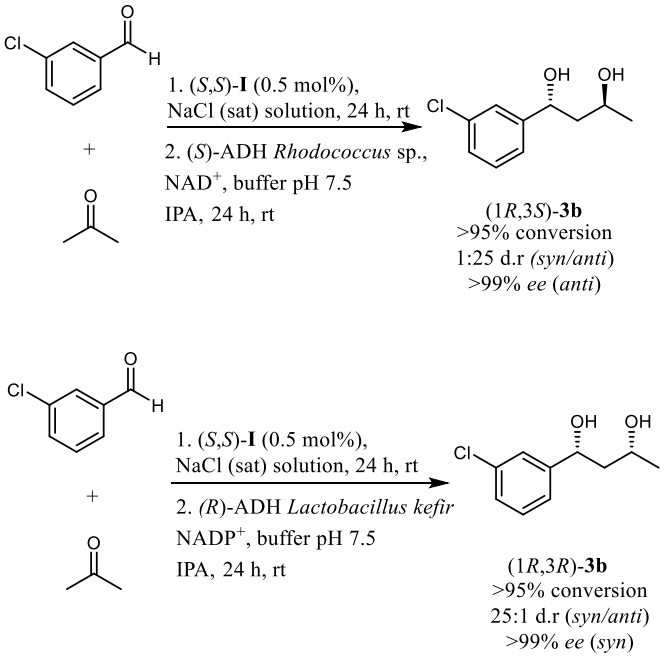
Scheme 3. One-pot organocatalysed aldol reaction combined with biocatalysed reduction to yield the optically active diol (1*R*,3*S*)-**3a**.[18]

Noteworthy, the generation of stereocentres is controlled only by the external catalyst, with marginal (if any) influence of the other chiral centre already present in the reactant. Thus, each stereo-centre is possible to be defined enantioselectively by the sole modulation of the organo- and the biocatalysts.

Encouraged for these initial results, the expansion of substrate scope to *meta*-substituted aromatic aldehydes was attempted, as shown in Scheme 4.[20] Similarly, asequential one-pot multistep system was set up using *m*-chlorobenzaldehyde as electrophile and 9 equivalents of acetone. Thus, with the chemoenzymatic one-pot synthesis as final goal, the organocatalysed aldol reaction was optimised in saturated of NaCl aqueous solution. As expected, catalyst loading plays a critical role in the stereochemical outcome of the reaction and a significant decrease in enantioselectivity was observed using 5 mol% of (*S*,*S*)-**I** after 48 h. Hence, different catalyst loading were tested, resulting in a high enantioselectivity at 0.5 mol%, while total reaction inhibition was shown at 10 mol% of **I**. The authors attributed this fact to a switch of reaction control from a kinetic to a thermodynamic regime.

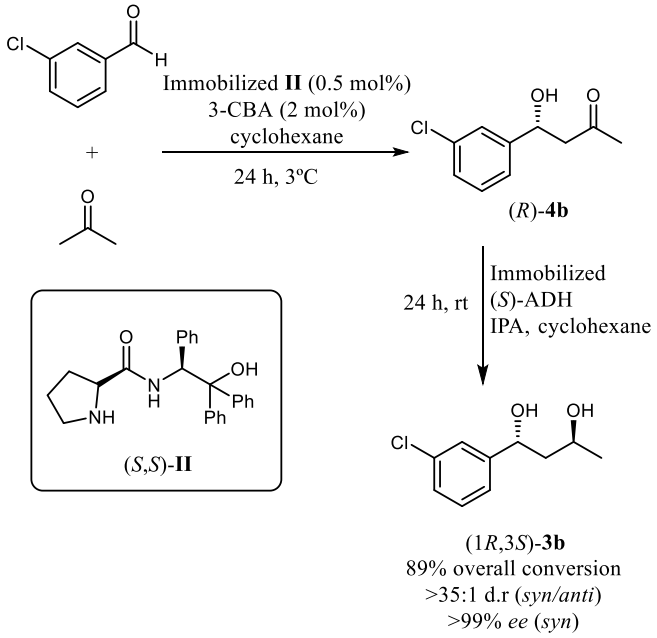
Thus, the combination of the organocatalysed aldol reaction with bioreduction using either (*R*)-ADH from *Rhodococcus* sp. or (*S*)-ADH from *Lactobacillus kefir* under optimised conditions, enabled the preparation of the corresponding

optically active 1,3-diols **3b** starting from *m*-chlorobenzaldehyde and acetone in one-pot two-step fashion in 48 h. The products were obtained with >95% of overall conversion, >25:1 of diastereomeric ratio and >99% of enantiomeric excess. As already mentioned, each catalyst or condition must be optimised according to the required performance. In this case, different amounts of enzyme were used depending on the activity of a catalyst towards the substrate, employing 10 units/mmol or 480 units/mmol with of (*S*)- or (*R*)-ADH, respectively.



Scheme 4. Synthesis of optically active 1,3-diols by combining organocatalysed aldol reaction in presence of (*S*,*S*)-**I** with bioreduction catalysed by (*S*)- or (*R*)-ADH in a sequential one-pot process to yield optically active diols (1*R*,3*S*)-**3b** and (1*R*,3*R*)-**3b**.[20]

obtained using a (*S*)-ADH from *Rhodococcus* sp. coimmobilised with its cofactor (NAD^{+}) onto the superabsorbent polymer Favor SXM 9155 (Evonik Industries AG).[23] The desired diol was obtained with high conversion (89%) and excellent diastereo- and enantioselectivity (*d.r.* >35:1, >99% *ee*). Moreover, the free proline-derivative catalyst **II** was also tested in this one-pot process and the same results were obtained in terms of activity and selectivity, making also suitable the combination in a one-pot process of a non-immobilised organocatalyst with an immobilised ADH in organic media.



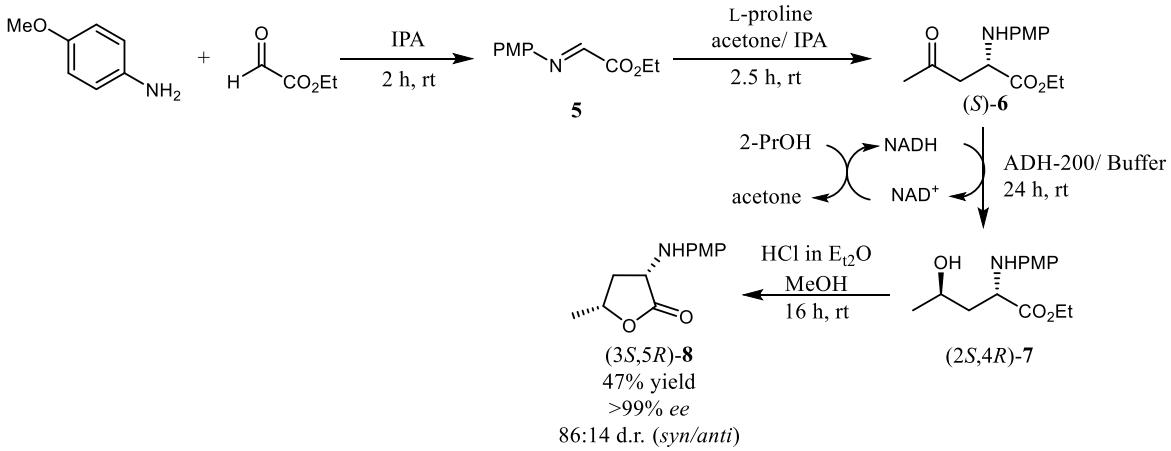
Scheme 5. Synthesis of (1*R*,3*S*)-**3b** in a one-pot procedure using both coimmobilised organocatalyst (*S*,*S*)-**II** and (*S*)-ADH from *Rhodococcus* sp. with its cofactor NAD+ in organic solvent medium.[21]

Very recently, by taking advantage of this sequential process, it was demonstrated the use of organic media in a similar one-pot process for an alternative preparation of chiral 1,3-diols, as shown in Scheme 5.[21] Immobilised organocatalyst **II** in acrylic polymeric beads catalysed the aldol reaction between aromatic aldehydes and acetone in organic media with similar results than those obtained with the free catalyst in aqueous media.[22] Likewise, employing immobilised **II**, it was possible to achieve the (*R*)-β-hydroxyketone (*R*)-**4a** starting from *m*-chlorobenzaldehyde (1,5 M) as acceptor with 95% of overall conversion and 95% *ee* after 24 h. However, 3-chlorobenzoic acid (3-CBA, 2 mol%) was needed as cocatalyst in order to overcome the low activity of (*S*,*S*)-**II** in non-aqueous conditions, and optimal performances were found at 3°C in order to preserve a suitable stereorecognition in an organic solvent such cyclohexane.
The immobilised catalyst was recycled by decanting and evaporating the organic layer before the bioreduction step. Likewise, chiral (1*S*,3*R*)-**3b** was

α-Amino-γ-butyrolactones are valuable building blocks in a set of natural compounds and pharmaceuticals. These compounds can be prepared by combining an L-proline-catalysed Mannich reaction to obtain an aminoketoester, which can be further reduced to any diastereomer alcohol by choosing the suitable ADH.[24] The resulting aminoalcohols cyclise, either spontaneously or under transesterification conditions (HCl-MeOH), to the desired aminolactones. The described synthesis comprises isolation of Mannich aduct intermediates, but the authors have also developed the one-pot reaction.
As shown in Scheme 6, the starting aldimine **5** is synthesised by mixing *p*-anisidine (0.53 mmol) with ethyl glyoxylate (0.55 mmol) in the presence of IPA (2.5 mL), which also serves as the solvent for the Mannich reaction catalysed by L-proline (0.13 mmol), and as hydrogen donor (cosubstrate) for the reduction of the formed ketone (*S*)-**6** to the corresponding aminoalcohol (2*S*,4*R*)-**7** in presence of the ADH-200, after dilution of the reaction medium with aqueous

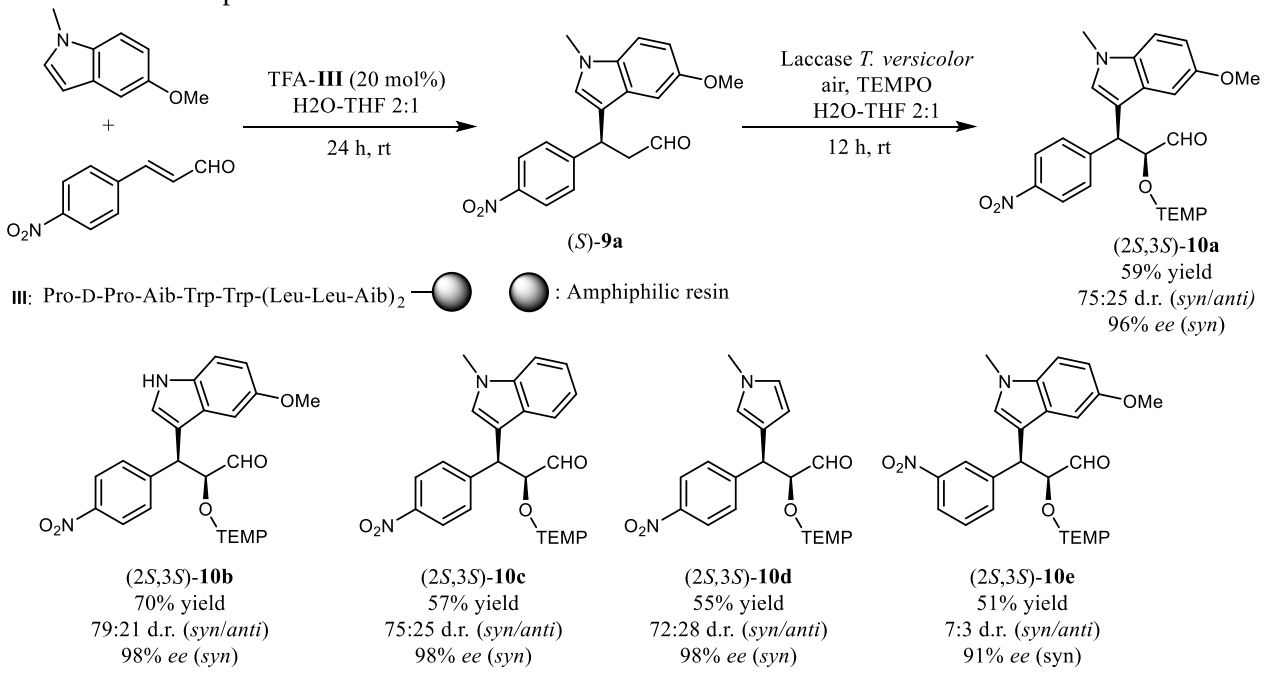
buffer. The final transesterification in presence of HCl in MeOH led to the enantiopure *syn*-(3*S*,5*R*)-lactone **8** with a 47% yield and diastereomeric excess of 72%. This value, lower to that obtained in the step-by-step synthesis, can be due to the 2-propanol employed as the solvent for the organocatalysed reaction, which might impair the selectivity of this step.

In a similar one-pot approach, aldimine **5** (1.0 mmol) was employed in the Mannich reaction with acetone (5 mL) as solvent and L-proline (0.25 mmol) as organocatalyst. After 16 h reaction, solvent was evaporated and ketone (*S*)-**6** was selectively reduced in presence of ADH-200 in buffer/IPA to aminoalcohol (2*S*,4*R*)-**7**. Further hydrolysis and purification afforded aminolactone (3*S*,5*R*)-**8** with 51% yield.



Scheme 6. Synthesis of aminolactone (3*S*,5*R*)-**8** in a one-pot procedure by combing a L-proline-catalysed Mannich reaction with a biocatalysed reduction of the formed ketone (*S*)-**6**.[24]

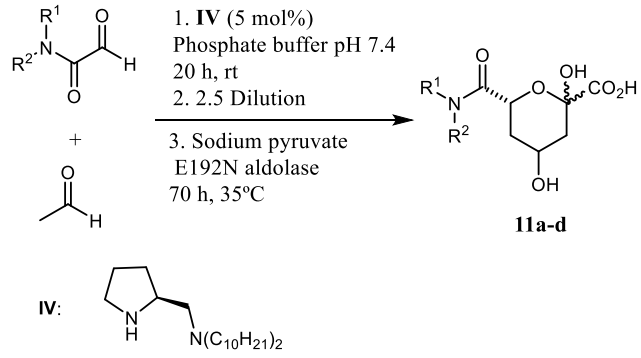
Resin-supported peptides can be employed in organocatalysed reactions carried out in aqueous environment.[25] These catalysts have been also applied to the one-pot sequential synthesis of oxyfunctionalised indoles,[26] in combination with the laccase from *Trametes versicolor*.[27] The resin-supported peptide **III** catalysed the asymmetric Friedel-Crafts alkylation of the starting indole (0.1 mmol) with an α,β-unsaturated aldehyde (0.15 mmol), through an iminium intermediate, while the laccase was able to catalyse the selective α-oxyamination of the aldehyde (*S*)-**9a** formed in the first step, as shown in Scheme 7. The one-pot reaction was carried out in aqueous medium. After 24 h for the Friedel-Crafts alkylation at room temperature, laccase and TEMPO (2,2,6,6-tetramethylpiperidin-1-oxyl radical) as mediator were added to the reaction mixture and stirred for additional 24 h. When employing 4-nitrocinnamaldehyde, the final product (2*S*,3*S*)-**10a** was recovered with a *syn*/*anti* ratio 75:25 and a 96% *ee* for the major diastereomer, but unfortunately with a very low conversion. The use of THF as reaction cosolvent (1:2 *ratio* with water) allowed increasing the substrates solubility, reaching a 59% yield for the final product with the same optical purity.



Scheme 7. One-pot synthesis of indole derivatives **10a-e** by combining a resin-supported peptide-catalysed Friedel-Crafts alkylation with a laccase-catalysed α-oxyamination.[26]

The reaction was then extended to other substituted indoles, to pyrrole and to 3-nitrocinnamaldehye. For all the examples, the *syn*/*anti* ratio of the final compounds (2*S*,3*S*)-**10b-e** were around 75:25, reaching excellent optical purities for the *syn* diastereomer, while yields were between 51 and 70%.

Table 1. One-pot three-component reaction catalysed by proline derivative **IV** and E192N NAL.[28]



Product	R^1	R^2	Yield [%][a)]	*cis/trans*[b)]
11a	Pr	Pr	40	78:22
11b	Me	Pr	48	57:43
11c	Me	Me	43	71:29
11d	Et	Et	51	61:39

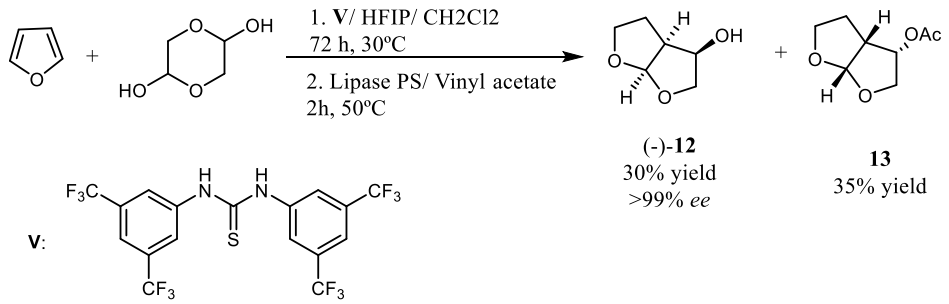
[a)] Based on glyoxylamide starting material. [b)] Determined by ^{1}H-NMR.

In 2012 it was described the first one-pot sequential three-component reaction in which two C-C bonds were created.[28] In the first step, a diamine organocatalyst **IV**(10 mol%) was able to perform the aldol reaction between a glyoxylamide (0.1 mmol) and acetaldehyde (0.1 mmol). The second C-C bond formation was carried out by the E192N mutant of *N*-acetylneuraminic acid lyase (NAL), which catalysed the aldol condensation between the aldehyde formed in the first step and pyruvate (0.1 mmol). This linear product spontaneously cyclises to achieve the hemiketalic final products **11a-d**.

As the enzymatic reaction has to be carried out in aqueous buffer, the organocatalysed aldol condensation was studied in this reaction medium. It was observed that with 10 mol% **IV**, reaction took place, but unfortunately with a modest diastereoselectivity and a drastic decrease in the enzymatic activity.

For this reason, the one-pot three-component reaction was carried out in a reaction medium that was diluted with aqueous buffer after the first aldol reaction. Thus, a set of glyoxylamides reacted with 10 equivalents of acetaldehyde in presence of 5 mol% organocatalyst in buffer pH 7.4 for 20 hours. After this time, a 2.5 dilution with buffer was performed and sodium pyruvate and E192N NAL were added. Reactions were shaken for other 70 h at 35ºC. Final products **11a-d** were recovered with yields around 40-51% and modest diastereoselectivities (Table 1). These lower values can be explained by two effects: 1) the absence of selectivity in the organocatalysed step; and 2) the low selectivity of the NAL-catalysed reaction.

A one-pot sequential organo- and biocatalysed process has been recently developed for the preparation of (3*R*,3*aS*,6*aR*)-hexahydrofuro[2,3-*b*]furan-3-ol (–)-**12** (bis-THF-alcohol, (Scheme 8), a structural motif of different HIV-1 protease inhibitors as Darunavir and Brecanavir. Firstly, the organocatalysed condensation of 1,2-dihydrofuran and glycoaldehyde dimerwas studied in order to obtain a mixture of *syn* and *anti* diastereomers of bis-THF-alcohol.[29] Reactions were carried out in presence of hexafluoroisopropyl alcohol (HFIP) as it has been described that this additive could increase the condensation yield by favouring the formation of glyceraldehyde monomer from its dimer. Among all the catalysts tested, Schreiner's thiourea (**V**) was the mostactive,[30] leading to a mixture of both *anti*-bis-THF-alcohol (the desired one to complete the synthesis) and the *syn* diastereomer with moderate yields.



Scheme 8. One- pot synthesis of the valuable synthon bis-THF-alcohol **12** by combining the Schreiner's thiourea **V** with lipase PS.[29]

Process optimization allowed obtaining a 70% yield of *anti*-**12** by using 2 equivalents of 1,2-dihydrofuran and HFIP, 1 mol% of Schreiner's catalysts in dichloromethane at 30ºC. The catalysed acetylation of the *anti*-alcohol with vinyl acetate at 50ºC was tested in presence of different lipases. Best results were achieved with lipase PS, recovering enantiopure *anti*-**12** with a 37% yield. Once the two catalytic processes have been optimised, the one-pot reaction was performed, first by carrying out the organo-catalysed condensation between 1.0 mmol of dimer and 4.0 mmol of 1,2-dihydrofuran in 2 mL of CH_2Cl_2 in presence of 4.0 mmol of HFIP and 2 mol% of thiourea **V** at 30ºC during 72 hours. After this time, vinyl acetate (2 mL) and lipase PS (260 mg) were added and the system was stirred at 50ºC for 24 h. After the crude purification, enantiopure *anti*-alcohol was obtained with 30% yield, while the *syn*-acetylated alcohol **13** was obtained with 35% yield. Scaling up of the process up to gram-scale (20 mmol of starting material) led to the same yield for the desired alcohol with 97% *ee*, demonstrating that this one-pot procedure can be practical for the preparation of pharmaceuticals.

3 Simultaneous one-pot processes combining organocatalysts and biocatalysts.

Apart from sequential one-pot reactions combining both organo- and biocatalysts, other designs have been performed in which all the reaction components are added at the beginning of the reaction (Figure 2).

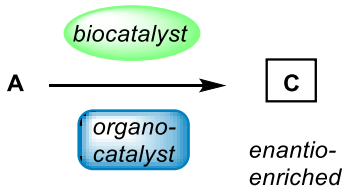
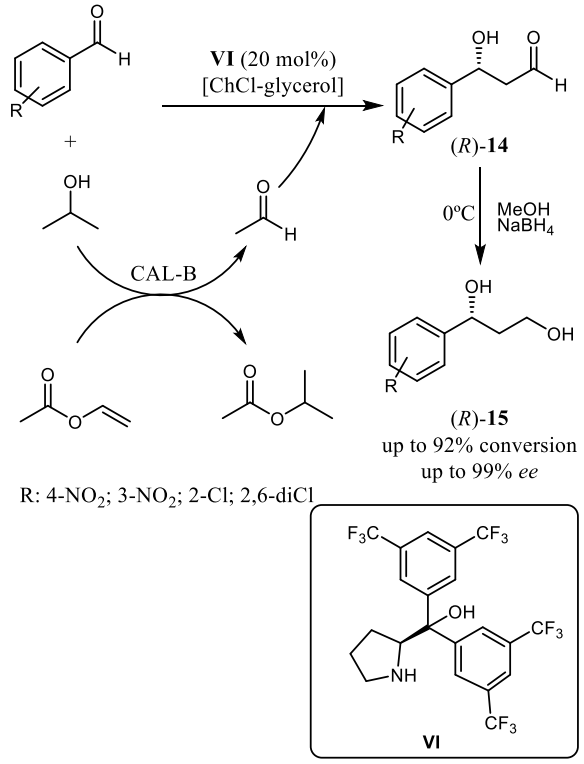


Figure 2. Typical simultaneous one-pot synthesis using organo- and biocatalysts.

Synthesis of diol (1*R*,3*S*)-**3b**(Scheme 4) has been recently performed by combining organocatalyst (*S*,*S*)-**I** and (*S*)-ADH from *Rhodococcus sp*. in a simultaneous way by adding both the organocatalyst and the biocatalyst at the beginning of the reaction.[31] Conditions of both the aldol reaction and the enzymatic reduction were optimised in order to develop an efficient process and to minimise possible side reactions as the biocatalysed reduction of the aldehyde, the biooxidation of diol **3b** to ketone **4b**, or aldol condensations. By employing an aldehyde concentration of 500 mM, 5 equiv. of acetone and 2-propanol as cosubstrate for the ADH (28% v v^{-1}), enantiopure (1*R*,3*S*)-**3b** can be obtained in a 60% conversion after 24 hours. Scaling up to 10 mmol of 3-chlorobenzaldehyde led to 50% conversion of the desired diol.

In 2014, an aldol reaction using an alternative source of aldehydes has been explored. In this line, it has been proposed the use of vinyl esters combined with lipases to generate acetaldehyde that will be employed in the organocatalysed synthesis of chiral 1,3-diols.[32] This strategy allows keeping low the acetaldehyde concentration, therefore diminishing the deleterious effects of this compound towards the enzymatic catalyst and avoiding self-condensation of this aldehyde. The lipase-catalysed transesterification of vinyl acetate with 2-propanol leads to the formation of acetaldehyde, which will serve as substrate in an organocatalysed aldol reaction, leading to chiral β-hydroxyaldehydes (*R*)-**14**. All the reactions occurred in a simultaneous one-pot fashion. Moreover, this process can be coupled with a further sequential reduction of the aldehyde to the corresponding (*R*)-1,3-diol (**15**) in presence of $NaBH_4$ (Scheme 9), minimising the retro-aldol reaction.
Thus, the one-pot reactions were performed using 1.0 mmol of aldehyde, 3.0 mmol of both vinyl acetate and propanol and 20 mol% of trifluoromethyl-substituted diphenylprolinol (**VI**) as organocatalyst. The deep eutectic solvent (DES) choline chloride-glycerol (1:2; 1.0 mL) was employed as reaction media,[33] while immobilised lipase B from *Candida antarctica* (CAL-B) carried out the acetaldehyde generation. Conversions were very high after 48 h (up to 92%), with reasonable high yields (up to 70%), and excellent enantioselectivities (up to >99%*ee*). Substrates with withdrawing substituents at any position of the aromatic ring were tested with similarly good results.

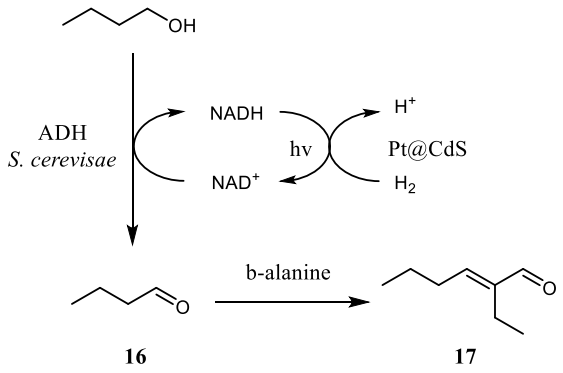


Scheme 9. Synthesis of (*R*)-1,3-diols in one-pot process combining CAL-B and trifluoromethyl prolinol catalyst **VI**in DES.[32]

The reaction was also extended to cinnamaldehyde derivatives such as α-bromocinnamaldehyde, with excellent *ee* (96%), although poor yield (14%). In addition, the use of aldehydes lacking electron-withdrawing groups such as 4-pyridinecarboxaldehyde or cinnamaldehyde led to disappointingly low yields (up to 2%) and enantioselectivity was fully suppressed. Furthermore, mandatory use of acetaldehyde leads always to β-hydroxyaldehydes, therefore chiral 1,3-diols are obtained with only one stereocentre. In this case, the stereoselectivity is totally controlled by the organocatalyst. DES and CAL-B could be reused up to six cycles without loss of enzymatic activity. Regarding the organocatalyst, yields were stable when fresh **VI** was added to the reaction medium while a slight loss of activity was observed in absence of extra organocatalyst.

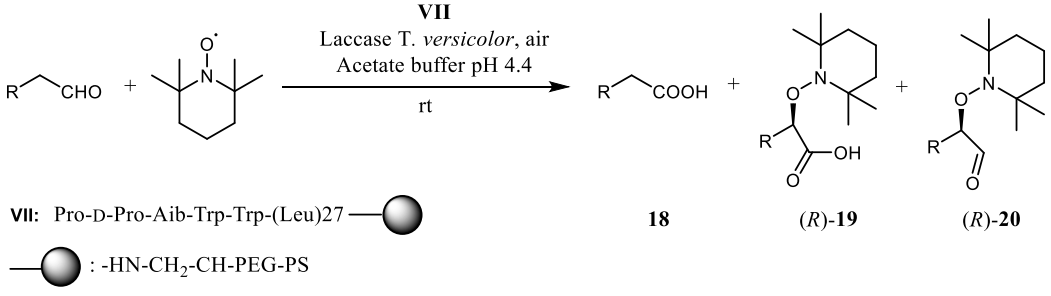
Combination of photo-, organo- and biocatalysis has recently allowed the conversion of *n*-butanol to 2-ethylhexenal (Scheme 10). In this simultaneous one-pot process the ADH from *Saccharomyces cerevisae* catalysed the oxidation of *n*-butanol to n-butyraldehyde (**16**) using NAD^+ as cofactor. The photocatalyst platinium-seeded cadmium sulfide (Pt@CdS) was employed for the cofactor regeneration. *n*-Butyraldehyde was converted to 2-ethylhexenal (**17**) in an aldol condensation catalysed by β–alanine.[34] The single reaction was performed by mixing 50 mM of *n*-butanol with 25 mM Pt@CdS, 3 mM NAD^+ and 550 mM by β–alanine. After degassing this system for one hour, 100 units of the ADH were added to initiate the reaction. The system was photoirradiated for 3 hours and the crude was extracted and analysed by ^{1}H NMR, being recovered 1.5 mM of **16** and 1.8 mM of **17**.

The one-pot system was also carried out in a synthetic acetone-butanol-ethanol (ABE) solution, using 15 mM acetone, 30 mM butanol and 5 mM ethanol. The three catalysts and NAD^+ were added to this solution. After 3 h photoirradation, the solution was extracted and analysed by ^{1}H NMR. A mixture of 1.0 mM of acetaldehyde, 1.0 of butyraldehyde and 1.5 mM of 2-ethylhexenal was recovered.



Scheme 10. Synthesis of 2-ethylhexenal **17** by combining three catalysts in a one-pot simultaneous procedure.[34]

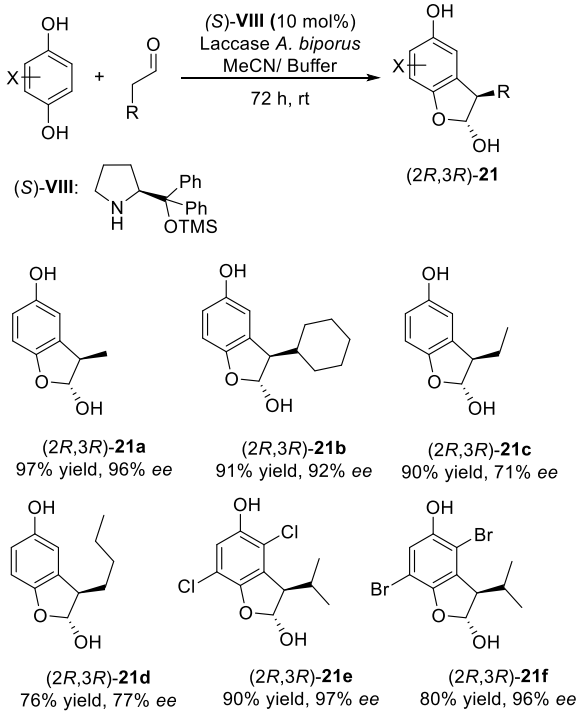
Resin-supported peptides (see Scheme 7) have been also employed in the simultaneous asymmetric α-oxyamination of aldehydes in combination with the laccase from *Trametes versicolor* and TEMPO as mediator.[35] This reaction can be regarded as a formal enolate/enamine one-electron oxidation,[36] and further radical trapping by TEMPO. Initial studies were devoted to analyse the products outcome at different reaction conditions. Thus, the laccase catalysed oxidation of 3-phenylpropanal in acetate buffer afforded the carboxylic acid (**18**) as sole product. When this biocatalysed reaction was carried out in presence of pyrrolidine as base catalyst, the racemic α-oxyaminated carboxylic acid (**19**) was formed together with α-unsubstituted carboxylic acid. The pyrrolidine-catalysed reaction performed in a water/1,4-dioxane mixture led to the formation of α-unsubstituted carboxylic acid and two racemic oxyaminated products, the chiral aldehyde (*R*)-**20** and the carboxylic acid (*R*)-**19**. The use of the resin-supported peptide **VII** as catalyst (Scheme 11) in water allowed the formation of (*R*)-α-oxyaminated carboxylic acid (65% conversion) with a 63% *ee* along with a smaller amount of **18** (35%). In view of these results, the one-pot reaction in presence of **VII** and the enzymatic system was extended to 4-arylbutanals. Thus, oxidation of 4-phenylbutanal led only to the α-oxyaminated aldehyde with a 71% yield and 82% *ee*. Similarly, complete conversion (53% yield) and 80% *ee* was achieved in the reaction of 4-(4-methoxyphenyl)butanal. This yield could be improved by performing the reaction in acetate buffer (74%), while the addition of the surfactant Tween 80 to the reaction mixture after 2 hours resulted in the α-oxyaminated carboxylic acid as major product (81%) with 64% yield and 91% *ee*.



Scheme 11. Peptide supported **VII**/laccase catalysed simultaneous one-pot α-oxyamination of aldehydes to obtain the orresponding chiral (*R*)-α-oxyaminated carboxylic acids (**19**) and aldehydes (**20**).[35]

Taking into account these encouraging results, the asymmetric oxidation of different aldehydes with laccase and **VII** was performed in absence and in presence of Tween 80. Reactions were carried out with 0.05 mmol of aldehyde and TEMPO, while using 0.01 mmol of **VII** and 0.5 mg of laccase in 0.5 mL of acetate buffer. In absence of surfactant, the oxyamination afforded the chiral aldehydes (*R*)-**20** in 1 h with good to moderate yields and optical purities close to 90%, while the presence of Tween 80 (1.0 μL) led to the carboxylic acids (*R*)-**19** with good yields and high enantioselectivities after 5-8 h. The system resulted highly efficient, being possible to reduce the amount of both catalysts to 5 mol% with negligible loss of activity and selectivity.

A set of 3-substituted-2,3-dihydrobenzofuran-2,5-diols (**21a-f**) were synthesised in a one-pot cascade procedure by combining an initial laccase-catalysed oxidation of 1,4-dihidroxybenzenes (hydroquinone derivatives), with the sequential aminocatalysed α-arylation of aldehydes, as shown in Scheme 12.[37] The reaction features laccase-catalysed oxidation of hydroquinone reagent giving rise to radical species that further add to the chiral enamine double bond. Reaction optimization was performed on the reaction between 3-methylbutanal and 1,4-dihidroxybenzene in presence of (*S*)-2-[diphenyl(trimethylsilyloxy)-methyl]pyrrolidine (*S*)-**VIII** as organocatalyst and the laccase from *Agaricus biporus*. Initial reaction was carried out with 0.25 mmol of 1,4-dihydroxybenzene in 0.5 mL buffer/acetonitrile pH 6.0 containing 10 mol% of (*S*)-**VIII**, 15 units of laccase and 5 equivalents of aldehyde per equivalent of 1,4-dihydroxybenzene.



Scheme 12. One-pot synthesis of chiral 3-substituted 2,3-dihydrobenzafuran-2,5-diols (**21a-f**) employing the secondary amine (*S*)-**VIII** as organocatalyst and laccase from *Agaricus biporus*.[37]

After 72 hours, the final product **21a** was obtained with 76% yield and 87% *ee*. The use of 45 units of laccase had a really positive effect on the procedure, as the yield increased up to 97% yield in the same reaction time with a 96% *ee*. The amount of organocatalyst had no effect on the cascade system. Reaction was scaled up to 2 mmol of 1,4-dihydroxybenzene, recovering the final hemiacetal with 84% yield and 92% *ee*. Another β-branched aldehyde as 2-cyclohexylacetaldehyde has been successfully tested in this procedure (91% yield and 92% *ee* for **21b**). On the contrary, unbranched aldehydes (for instance **21c-d**) led to moderate to good yields (51% to 90%) and lower optical purities, being observed that shorter reaction times afforded higher selectivities. Studies on this line suggested that the final product tautomerises, and thus the aldehyde forms an iminium ion with the aminocatalyst, leading to enamine after deprotonation, which upon hydrolysis afforded again the hemiacetal. As branched aldehydes are more stable in the hemiacetalic form, this racemisation did not occur, while a significant amount of aldehyde is observed for the unbranched ones, which induces this decrease in the optical purity. The use of substituted 1,4-dihydroxybenzes as the 2,6-dichloro- (**21e**) or the 2,6-dibromo (**21f**) derivatives in the reaction with 3-methylbutanal allowed to obtain the final products with good yields (around 90%) and excellent optical purities (around 95%).

Chiral functionalised cyclopentenones have been employed as precursors of natural products and biologically active compounds. Different methodologies have been proposed to synthesise them, but all suffered from a number of drawbacks. Therefore, a one-pot approach starting from a pyranone to achieve the optically active cyclo-pentenone has been recently developed. This methodology consisted in a sequential process in which an initial organocatalysed rearrangement converts the pyranone into a cyclopentenone presenting a hydroxyl moiety **22a**, which then underwent a lipase-catalysed kinetic resolution to yield the alcohol (+)-**22a** and the ester (–)-**23a** (Scheme 13a).[38] The rearrangement was optimised in order to have compatible conditions with the enzymatic reaction and to prevent the decomposition of the obtained cyclopentanone into the undesired enone (**24a**). Thus, after testing different reaction parameters, it was possible to obtain **23a** in 85% yield, with only 3% of enone side product, after a 24

h treatment of the starting material with 0.15 equivalents of the amine 4-diazabicyclo[2.2.2]octane (DABCO) at 50ºC in presence of *tert*-butanol as solvent. When combining the rearrangement at the optimal conditions with the enzymatic resolution of the alcohol formed in presence of vinyl acetate, several lipases were tested, being achieved the best results with lipase AK. Starting from 100 mg of pyranone, 5 equiv. of vinyl acetate, 30 mol% DABCO and 50 mg of lipase in 1.0 mK of *tert*-butanol led to chiral acetate (–)-**23a** was isolated with 55% yield and 80% *ee* after 10 days, while the starting alcohol (+)-**22a** was recovered in 35% yield and only 11% *ee*. The low optical purity of the alcohol can be explained by its racemisation at the reaction conditions, which open up the opportunity for developing a dynamic kinetic resolution (DKR) in order to obtain the acylated product **23a** with a high yield and optical purity (see Figure 3).[39]

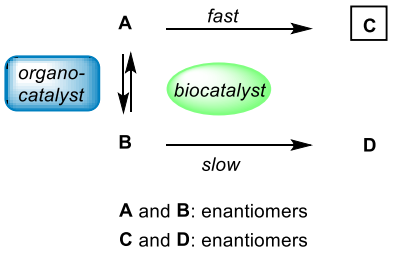
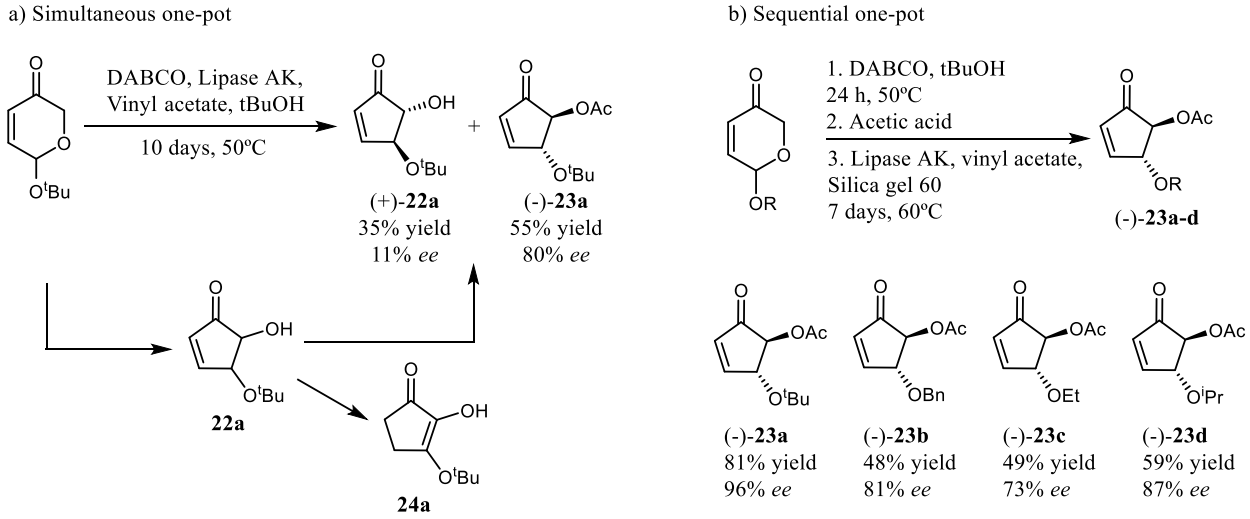


Figure 3. General representation for a dynamic kinetic resolution (DKR).

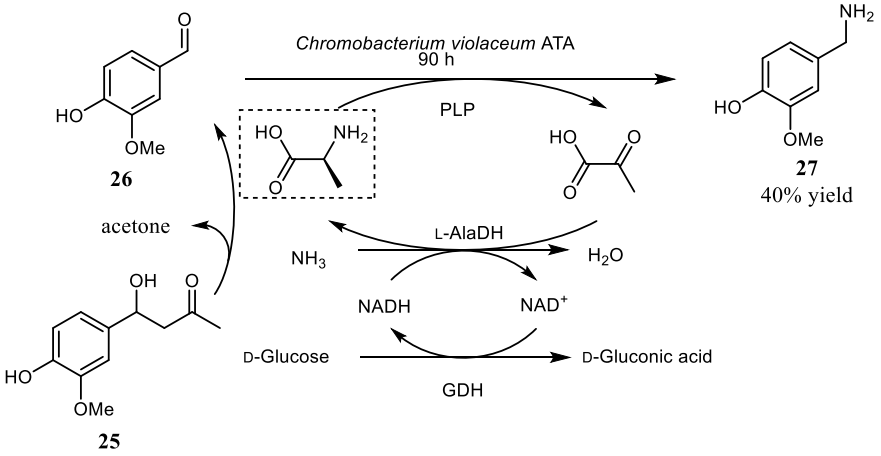
Thus, alcohol (+)-**22a** racemisation was induced in presence of strong acidic medium (pH 1.0-2.0) in the reaction mixture, but these conditions have a negative effect of lipase activity. Racemisation is very effective at the acylation conditions, which can be likely due to the enzymatic activity or to the acetic acid production by vinyl acetate hydrolysis. It was also observed that racemisation rate was improved by using silica gel 60 (5mg/mg of lipase AK), which also present acidic character. In order to combine in a one-pot process the conditions for the racemisation and the biocatalysed acetylation, a sequential one-pot procedure was developed (Scheme 13b).[40].The pyranone (2.35 mmol) rearrangement was carried out during 24 h with DABCO (0.36 mmol) in ^{t}BuOH (2.0 mL), after which the mixture was neutralised with acetic acid. Then, lipase AK (400 mg), silica gel 60 (2.0 g) and vinyl acetate (5.0 equiv.) were added and reacted for 7 days in order to achieve the acetyl cyclopentenone (–)-**23a** with 81% yield and 95% *ee*, while the hydroxyl cyclopentenone was recovered with only 10% *ee* and a yield of 4%. This methodology was then extended to other (–)-cyclopentenones (**23b-d**) but in all cases, lower yields and optical purities were obtained.



Scheme 13. a) One-pot simultaneous synthesis of chiral cyclopentenones (+)-**22a** and (–)-**23a** by DABCO-catalysed rearrangement and lipase AK catalysed kinetic resolution;[38] b) One-pot sequential synthesis and DKR of pyranones catalysed by DABCO and lipase AK for the preparation of acetylated cyclopentenones (–)-**23a-d**.[40]

A different approach to the organo- and biocatalysed one-pot systems has been recently described in the preparation of capsaicin analogues, compounds with high biological interest, starting from lignin-derived compounds using a multi-step procedure.[41] The last step of this synthesis is the biocatalysed acylation of vanillylamine (**27**). Authors have proposed different procedures for its preparation by combining different catalysts. One of the procedures consist in a simultaneous one-step process starting from 4-hydroxy-4-(4-hydroxy-3-methoxyphenyl)butan-2-one (**25**) in presence of L-alanine as organocatalyst and the aminotransferase from *Chromobacterium violaceum* as biocatalyst using aqueous buffer. L-Alanine catalysed the retro-aldol reaction of **25** to vanillin (**26**) and is also employed as amino donor in the biotransformation of vanillin to **27** catalysed by the aminotransferase. An enzyme cascade system was employed in order to regenerate the L-alalnine and to increase the reaction conversions, as shown in

Scheme 14. During **26** transamination, L-alanine is converted to L-pyruvate. In presence of L-alanine dehydrogenase (L-AlaDH), L-alanine is regenerated using ammonia as nitrogen source. As L-AlaDH is a NADH dependent enzyme, glucose dehydrogenase (GDH) is required to regenerate this cofactor by converting D-glucose into D-gluconic acid. The overall system to convert one equivalent of vanillin to vanillylamine requires one equivalent of ammonia and one equivalent of glucose. The combination of the organocatalyst (250 mM) and the enzymatic cascade system afforded vanillylamine from **25** (2.5 mM) substrate in HEPES Buffer (1.0 mL) with a complete conversion and a 40% yield after 90 hours. The remaining 60% yield corresponds to the dehydrated aldol condensation product, obtained in a side reaction also catalysed by L-alanine, as suggested by the authors.



Scheme 14. One-pot simultaneous synthesis of vanillylamine **27** employing L-alanine and aminotransferase from *Chromobacterium violaceum.*[41]

4 One-pot processes combining biocatalysts and non-traditional organic catalysts.

4.1 Reactions catalysed by enzymes and base catalysts

In 1991 it was reported the one-pot synthesis of optically active cyanohydrin acetates starting from aldehydes, by combining the anion exchange resin (OH- form) catalysed transcyanation of the aldehyde and cyanohydrin acetone and the lipase-catalysed kinetic resolution of the resulting cyanohydrin.[42] As the formation of the cyanohydrin, is a reversible process, this compound will suffer a fast racemisation. Thus, the enzymatic acylation will afford a single enantiomer of the cyanohydrin acetate with high yield in a DKR.

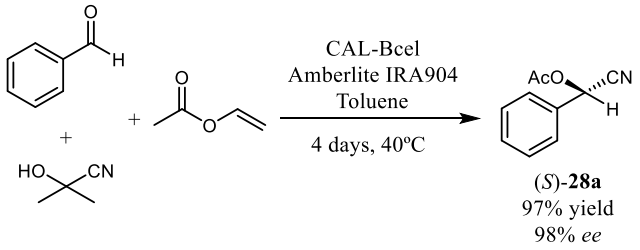
After analysing different anion-exchange resins, reactions were carried out with Amberlite IRA 904 and the lipase from *Pseudomonas sp*, M-12-33 from Amano, in presence of isopropenyl acetate as acyl donor. After long reaction times (2-6 days), it was possible to obtain the chiral (*S*)-cyanohydrin acetates with high yields and optical purities, except for the 1-naphthyl derivative.

This kind of reaction has been exploited for the synthesis of other chiral cyanohydrin acetates. In 2003, it was performed the preparation of optically active phenylfuran-based cyanohydrins esters, valuable building blocks of biologically active compounds, using Amberlite IRA 904 and lipase PS from *Pseudomonas cepacia*.[43] The basic exchange resin was able to perform the catalysed transcyanation between phenylfuranaldehydes and acetone cyanohydrin, while the cyanohydrins formed were selectively acylated in the presence of the biocatalyst and vinyl butanoate as acyl donor. The one-pot process afforded the final (*R*)-cyanohydrin esters with high yields and optical purities. The use of higher amounts of enzyme and temperatures led to much shorter reaction times, a result to be taken into account for the preparation of the desired products at gram-scale. The authors have also performed the synthesis of chiral (*R*)-benzothiazol-based cyanohydrin acetates by the same procedure.[44] For these products, the best results were achieved again with Amberlite IRA 904 combined with the lipase from *Candida antarctica* A immobilised on Celite, which led to the best selectivities in the acetylation of the cyanohydrins with vinyl acetate. The one-pot processes were carried out with high selectivities and complete conversion after 2-3 days using 10 mg mL^{-1} of lipase preparation. Lower amounts led to longer times, while an excess of enzyme afforded a decrease in the optical purity of the (*R*)-cyanohydrin acetates as the racemisation step was not fast enough when compared with the enzymatic reaction.

In 2002, the enantioselective synthesis of (*S*)-mandelonitrile acetate (**28a**) through a DKR was developed starting from benzaldehyde (0.8 mmol), acetone cyanohydrin (2 equiv.) and isopropenyl acetate (3 equiv.) in toluene (8.0 mL). Amberlite IRA904 (0.25 equiv., OH^- form) and CAL-B (80 mg)were used in this simultaneous one-pot three-step procedure.[45] Amberlite is able to perform the release of cyanide hydrogen from acetone cyanohydrin as

well as the HCN addition to benzaldehyde. Both processes are reversible, something essential for the preparation and racemisation of mandelonitrile, which rapidly occurred at either 40 or 60ºC. CAL-B catalysed acetylation of mandelonitrile at those temperatures is a very selective process, in which enantiopure (*S*)-**28a** was recovered. Unfortunately, when the reaction was carried out in a one-pot approach, it was observed that the kinetic resolution of mandelonitrile is performed without racemisation of the starting material, indicating that at these conditions the Amberlite resin is deactivated. This is likely due to the presence of water in the reaction medium, responsible for the hydrolysis of isopropenyl acetate to acetic acid, which neutralises the alkaline resin. For this reason, an extra amount of basic resin was added to the reaction medium, but enantiopure (*S*)-**28a** was obtained with low yield (16%) after 45 h.

A further development in order to circumvent this low yield was made by immobilising CAL-B on Celite (CAL-Bcel), a natural silicate able to adsorb water. The use of this biocatalyst preparation led to a 97% yield of almost enantiopure (*S*)-mandelonitrile acetate after 4 days reaction (Scheme 15).[46]



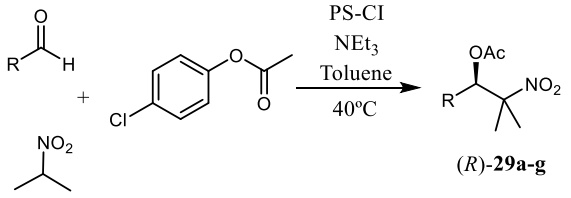
Scheme 15. One-pot three-step procedure for the synthesis of (*S*)-**28a** using CAL-B supported on Celite and the anion exchange resin Amberlite IRA904.[46]

The combination in a simultaneous one-pot DKR of an organocatalysed nitroaldol (Henry) reaction with a lipase-catalysed acylation has been used for the preparation of a set of (*R*)-β-nitroacetates (**29a-g**) with high yields and optical purities.[47] As the Henry reaction requires the control of the equilibrium, this reaction has been used for two purposes: 1) synthesis of the β-nitroalcohol, the substrate of the enzymatic acylation; and 2) racemization of the non-reactive β-nitroalcohol enantiomer.

Several organic bases have been tested as catalysts for the nitroaldol reaction, achieving the best results with triethylamine. Lipases from *Pseudomonas* led to the better performance in the acylation step. This process was optimised by employing 30 mg of *Pseudomonas cepacia* lipase preparation (PS-CI) per 0.05 mmol of substrate in toluene (1.0 mL) at room temperature in the presence of 5 equivalents of vinyl acetate. When the one-pot DKR was conducted with an excess of nitroalkane, 2 equivalents of triethylamine, 1 equivalent of *p*-nitrobenzaldehyde, 5 equivalents of vinyl acetate and PS-CI, a 65% yield of the corresponding β-nitroacetate was recovered with 85% enantiomeric excess. A significant amount of two byproducts was obtained. One of them is formed by the coupling of the acetaldehyde (generated as byproduct in the acylation reaction) with the nitroalkane, while the other one was achieved by the acylation of this first byproduct.

In order to improve the process yield, different acyl donors were tested in the PS-CI-catalysed acylation. Although *p*-chlorobenzyl acetate led to lower activities as compared with vinyl acetate, no side reactions were observed, so this compound was chosen for further development (Table 2). The simultaneous one-pot DKR starting from 2-nitropropane (0.5 mmol) and *p*-nitrobenzaldehyde (0.125 mmol) in presence of trimethylamine (0.25 mmol), PS-CI (90 mg) and *p*-chlorobenzyl acetate (106 mg) in toluene (0.25 mL) and molecular sieves (20 mg) at 40ºC afforded (*R*)-**28a** with 90% yield and complete selectivity. The biocatalyst can be recovered without loss of activity. This methodology was extended to other benzaldehydes by slightly modifying the reagents amounts, achieving excellent yields and selectivities for (*R*)-**28b-g**.

Table 2. Henry reaction and base-catalysed racemisation to obtain optically active (*R*)-β-nitroacetates in presence of *Pseudomonas cepacia* lipase.[47]



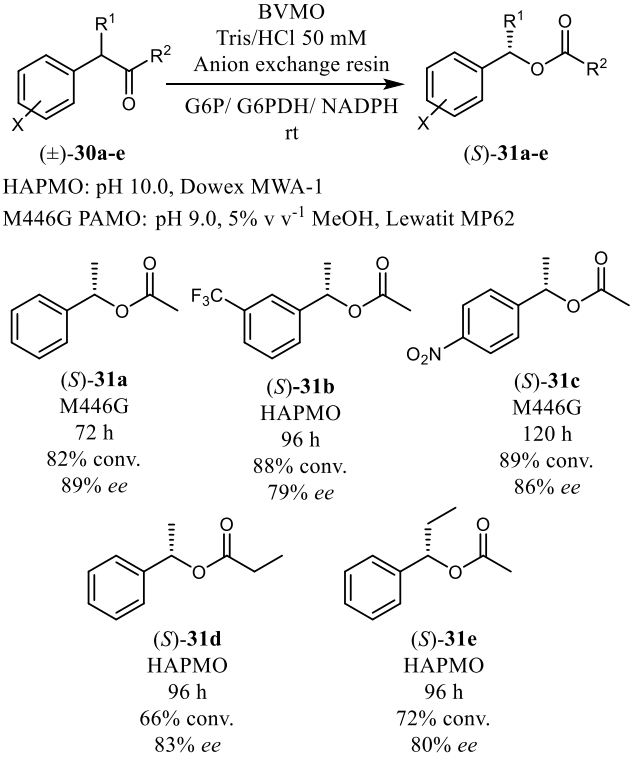
Product	R	t [days]	Yield [%][a)]	*ee* [%][b)]
29a	4-O_2N-Ph	2	90	99
29b	4-F_3C-Ph	3	89	97
29c	3-NC-Ph	3	92	91
29d	4-F-Ph	4	85	98
29e	4-Cl-Ph	4	83	97
29f	Ph	4	79	91
29g	4-MeO-Ph	4	28	99

[a)]Isolated yield.[b)]Determined by HPLC.

In the last few years, a number of examples have been reported regarding one-pot reactions in which an organic base-catalyst promotes the racemisation of the biocatalytic reaction substrate, in order to achieve DKRs.

The selective Baeyer-Villiger oxidation of racemic benzylketones (±)-**30a-e** (50 mg) catalysed by the Baeyer-Villiger monooxygenase (BVMO)[48] 4-hydroxyacetophenone monooxygenase (HAPMO)[49] from *Pseudomonas fluorescens* ACB (2.0 μM), using glucose 6-phosphate (G6P, 20 mM) and glucose-6-phosphate dehydrogenase (G6PDH, 50 units) as NADPH (0.2 mM) cofactor regeneration system, was combined with the racemisation of the starting

material in presence of the weak anion exchange resin Dowex MWA-1 (100 mg). By using this simultaneous one-pot process benzylesters (*S*)-**31a-e** were obtained with good yields and optical purities (Scheme 16).[50] The use of strong anion exchange resins allowed higher racemisation rates, whereas negatively affected the enzymatic system. Reactions were performed in Tris/HCl buffer pH 10.0 in order to ensure a racemisation rate higher than the conversion of the slower ketone enantiomer. This procedure was then extended to the Baeyer-Villiger oxidations catalysed by the M446G mutant of phenylacetone monooxygenase (PAMO) from *Thermobifida fusca*.[51] For this biocatalyst, the best results were achieved in Tris/HCl buffer pH 9.0 containing a 5% v v^{-1} as cosolvent while using the weak anion exchange resin Lewatit MP62.[52] The space-time yield of the reaction, expressed as mmoles of ketone consumed per hour and per liter of solution, increased up to 60. Higher substrate loading led to a decrease in this parameter, with no effect on the selectivity. Final (*S*)-benzylesters were recovered with moderate to good yields and optical purities between 65 and 86%.



Scheme 16. Dynamic kinetic resolution of racemic benzylketones (±)-**30a-e** catalysed by Baeyer-Villiger monooxygenases and weak anion exchange resins.

ADHs have been also tested in dynamic processes. These enzymes are valuable tools for the production of chiral alcohols with one or more stereocentrs. The racemisation of the non-reactive stereocentre is possible in order to obtain multiple chiral centres in only one process. The epimerizable chiral centre is located in an adjacent position to the carbonyl moiety, containing an acidic proton which facilitates the racemization, in the so-called dynamic reductive kinetic resolutions (DYRKRs).[53]

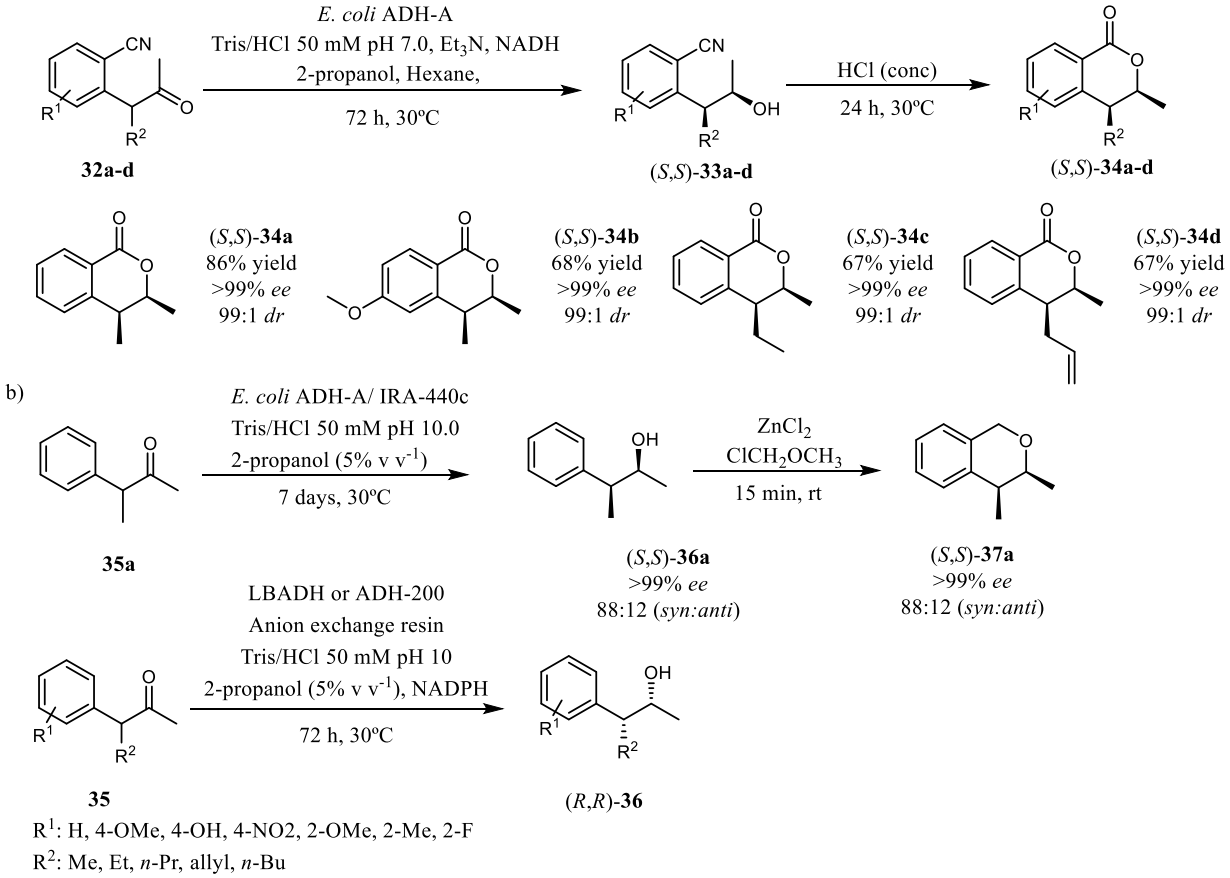
A DYRKR has been described for the preparation of chiral 3,4-dialkyl-3,4-dihydroisocoumarins **34** starting from 2-(3-oxoalkyl)benzonitriles **32** through a simultaneous one-pot biocatalytic reduction combined with substrate racemisation (Scheme 17a).[54] 2-(3-Oxobutan-2-yl)benzonitrile (±)-**32a** was chosen as model substrate, being reduced in presence of the Prelog alcohol dehydrogenase ADH-A from *Rhodococcus ruber* overexpressed in *E. coli*. The bioreduction carried out in Tris-HCl buffer pH 7.5 containing 5% v v^{-1} 2-propanol cosubstrate and 5% v v^{-1} hexane cosolvent and 30ºC afforded the chiral (*S*,*S*)-alcohol **33a** with an excellent enantio- and diastereoselectivity and a 56% conversion after 24 hours. As racemisation systems, two possible alternatives were studied: 1) triethylamine, or 2) anion exchange resin Dowex MWA-1. The DYRKR process was studied with both systems, achieving a slightly higher conversion with the anion exchange resin to obtain enantiopure (*S*,*S*)-**33a** with 86% conversion after 92 hours. This procedure was performed with other ketones bearing different substituents in the aromatic ring or in the stereocentre, leading to the final enantiopure alcohols with good to excellent yields and complete diastereoselectivity. The process was further extended by carrying out the one-pot acid-catalysed cyclization of the (*S*,*S*)-alcohols obtained by DYRKR after 72 hours, leading to the corresponding (*S*,*S*)-3-dialkyl-3,4-dihydroiso-coumarins **34** with good yields in most of cases while excellent enantio- and diastereoselectivity was observed. At the reaction conditions employed to obtain (*S*,*S*)-**34a-d**, starting from 0.3 mmol of **32a-d** in 13 mL Tris-HCl 50 mM phH 7.5 buffer, triethylamine (1% v v^{-1}) was chosen as racemisation reagent given its higher reliability against Dowex MWA-1.

A similar methodology has been recently applied for the DYRKR of 3-arylalkan-2-ones **35** to obtain the corresponding chiral substituted propan-2-ols **36**,[55] which are converted to isochromans **37**, valuable building blocks in organic synthesis (Scheme 17b). The initial tests with racemic (±)-3-phenylbutan-2-one **35a** in presence of the Prelog *E. coli*/ADH-A afforded (*S*,*S*)-**36a** with good selectivity values and conversions close to 50% at short reaction times (83:17 of diastereomeric ratio for the *syn* diastereomer). This compound is transformed into the desired isochroman (*S*,*S*)-**37a** by treatment with zinc chloride in chloromethoxymethane at room temperature.

Substrate racemisation was studied in presence of anion exchange resins as DowexMWA-1 and Amberlite IRA-440C. A high pH was required to reach an effective racemisation, however, at these conditions the enzyme suffers from severe inactivation. In order to circumvent this drawback, a higher reaction temperature was used (30ºC), while ADH-A was added stepwise at the reaction medium.

The optimal DYKRK conditions were applied over a set of racemic benzylketones (0.01 mmol). Bioreductions were carried out at 30ºC in Tris-HCl 50 mM pH 10.0 (0.35 mL) using one of the exchange resins, while ADH-A was added in portions over 3-4 days. Excellent enantio- and diastereoselectivities were observed for the C-2 substituted 3-arylbutan-2-ones. When Amberlite IRA-440C was employed, a significant decrease in the reactivity was observed for alkyl chains longer than methyl, while the use of Dowex MWA-1 led to better conversions, although with lower diastereomeric ratios.

Experiments were also performed with two anti-Prelog ADHs: Alcohol dehydrogenase from *Lactobacillus brevis* (LBADH) and ADH-200, which led to the corresponding (*R*,*R*)-propan-2-ols **36**. It was observed that Amberlite IRA-440C caused inhibition in both biocatalysts, so the dynamic processes were carried out in presence of Dowex MWA-1. For all the racemic benzylketones tested, moderate to good conversions were measured together with excellent selectivities and high diasteroselectivities for the (*R*,*R*)-alcohols obtained.

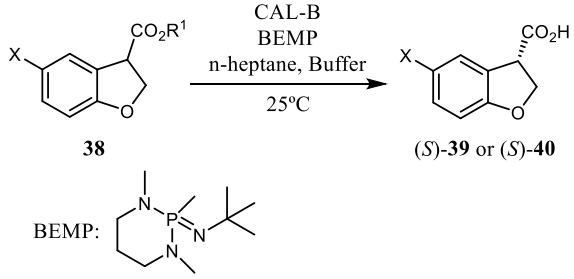


Scheme 17. a) DYRKR of 2-(3-oxoalkyl)benzonitriles to obtain dihydroisocoumarins (*S*,*S*)-**34a-d** using ADHs and trimethylamine as base catalyst;[54] b) DYRKR of 3-aryl-2-butanones to yield the corresponding chiral arylpropan-2-ols **36** and the isochroman (*S*,*S*)-**37a** by a one-pot process employing ADHs and an anion exchange resin.[55]

The chemoenzymatic synthesis of (*S*)-2,3-dihydrobenzo[*b*]furan-3-carboxylic acid (**39**) and (*S*)-5-chloro-2,3-dihidrobenzo[*b*]furan-3-carboxylic acid (**40**), valuable precursors in the synthesis of biologically active compounds, features as key step the sequential one-pot biocatalysed hydrolysis of its racemic methyl or ethyl esters (**38**) combined with the substrate racemisation in presence of an organic base.[56] Initial screening on the enzymes for the kinetic resolution were performed by an HPLC-CD selectivity assay, leading to CAL-B and *Bacillus subtilis* esterase (BS3) as the best biocatalysts for this process. For both enzymes, enantioselectivities were excellent, recovering both ester and acid with 50% conversion and yields higher than 46%. While CAL-B hydrolysed the (*S*)-enantiomer of the ester (4.4 mmol) to yield (*S*)-**39** and (*S*)-**40**, BS3 hydrolysed the (*R*)-antipode in enantiocomplementary fashion. Substrate racemisation was studied in presence of different organic bases. The Schwesinger base *tert*-butylimino-2-diethylamino-1,3-dimethylperhydro-1,2,3-diazophosphorine (BEMP) was the more convenient for this process.[57] Depending on the substrate structure, different amounts of BEMP were required to achieve a satisfactory racemisation. As BEMP can be inactivated in aqueous medium, a reaction setup for the one-pot reaction with separation of both processes employed. Thus, a flask was connected with a peristaltic pump to a column containing immobilised BEMP. The reaction was

carried out in a biphasic system buffer/*n*-heptane (60 mL, 2:1) in which the biocatalyst and the enantiopure acid product stand in the aqueous phase, while the ester (kept in the organic phase) was continuously pumped through the BEMP column. In this system, the ester is in a continuous racemisation while the acid accumulates in the aqueous phase. In order to avoid BEMP leaching, which can lead to side-reactions and biocatalyst inactivation, the base was protected at the column with a second layer of ion-exchange resin. As shown in Table 3, the use of this system at preparative scale (500 mg, 2.81 mmol of the corresponding ester) led to the chiral acids with high optical purities and good yields depending on the substrate structure.

Table 3. DKR of racemic esters in presence of CAL-B and BEMP as racemisation reagent to obtain the (*S*)-2,3-dihydrobenzo[*b*]furans **39** or **40**.[56]



Product	X	R^1	t [h]	BEMP [equiv]	Yield [%]	*ee* [%][a)]
39	H	Me	26	1.0	95	90
39	H	Me	24	1.5	92	>95
40	Cl	Me	24	2.0	71	>99
40	Cl	Et	24	1.0	82	>99

[a)] Measured by HPLC.

4.2 Enzymatic regeneration of a redox catalyst in a one-pot procedure.

A significant advance in the organocatalysed redox reactions has been experienced lately although it cannot be paralelled with the explosion of organocatalysed C-C bond forming reactions developed during the same period. Organocatalysed redox reactions are mostly focused on alkene epoxidations,[58] thioether sulfoxidations,[59] and Baeyer-Villiger oxidations[60] employing (a) chiral nitrosyl radical- dioxirane- or oxaziridine-based catalysts, among others. More recently, successful organocatalytic asymmetric alkene reductions under hydrogen transfer conditions have been reported.[61]
With this scenario and considering the vast knowledge on biocatalysed oxidations,[62] it is reasonable to conceive fruitful combinations of enzymatic regeneration of redox organocatalysts. Indeed, examples of such processes can be traced back to 1990.[63]

So far, two main strategies have been explored with a great deal of success (Figure 4): 1) the hydrolase-catalysed perhydrolysis of carboxylic acid/esters (the organocatalysts) giving rise to an organic peracid that performs as direct oxidizing agent and, 2) the versatile laccase/mediator system for several one- and two-electron oxidation processes.

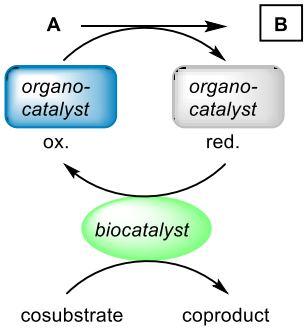
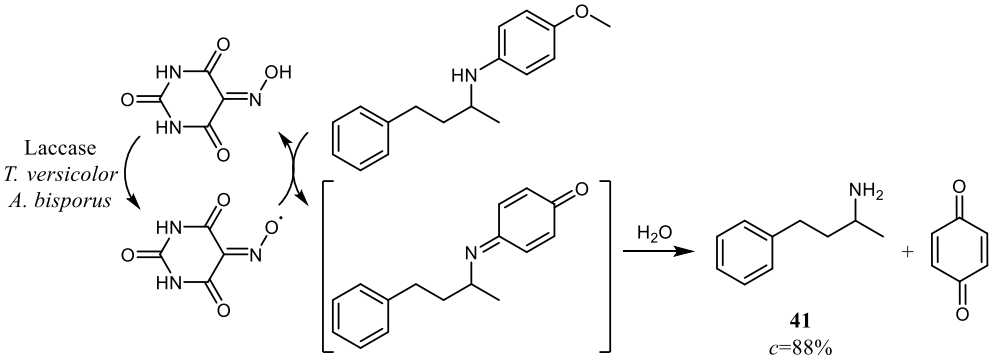


Figure 4. Biocatalysed regeneration of redox catalysts in one-pot fashion.

In this hydrolase/carboxylic acid one-pot procedure, substoichiometric quantities of carboxylic derivative can be employed that and after peracid formation, it shall work as oxo-transfer catalyst from hydrogen peroxide to the substrate (mostly ketones, olefins and sulfides).
The use of lipases in promiscuous reactions has been extensively reviewed in the last years.[64] One of those lipase-catalysed promiscuous processes is, as mentioned, the reaction between an ester or carboxylic acid with H_2O_2 to render the corresponding peracid. When this carboxylic derivative is used in substoichiometric quantity, a catalytic oxidation reaction can be coupled to the enzymatic perhydrolysis reaction.
Remarkably, already in 1990 it was reported that immobilised CAL B and octanoic acid (10 % mol) in the presence of a slight molar excess of H_2O_2 (added in portions), results in the epoxidation of several alkenes under solvent free conditions. The reactions were stopped at 15 h with almost full conversion in most cases.[63] Ten years later, a similar system was set up but taking place in ionic liquids, thus obtaining the cyclohexene oxide with high selectivity and 83% of conversion.[65]
As expected several reports dealt with lipase-catalysed generation of percarboxylic acid to carry out reactions other than epoxidation.[66] However, in the vast majority of these cases, the carboxylic acid (here considered as the organocatalyst) is added in overstoichiometric quantities, thus, the sulfoxidation or the Baeyer-Villiger ketone oxidation are not strictly catalytic and therefore fall out of this review.
On the other hand, laccases are able to oxidise substrates using O_2 as terminal electron acceptor. When the enzyme redox potential is not enough for the oxidation of a given substrate, chemist usually adopt a small organic molecule as an electron mediator, in the so-called 'laccase/mediator system' (LMS).[27] This clean reaction is readily employed for

the transient generation of highly reactive intermediates that can act as one- and two-electron shuttle between the substrate and O_2 that accept protons to form H_2O. In a wide sense, this organic mediator employed in catalytic quantities or its reactive intermediate can be regarded as the actual organic catalyst.[67] Furthermore, these mediators can act by different mechanisms, such as electron-transfer (ET), radical hydrogen-atom-transfer (HAT) or polar mechanism, depending on its electronic nature.

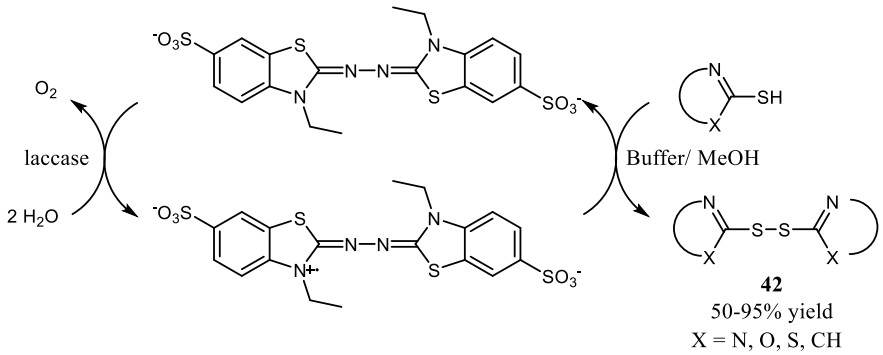
Several systems involving an oxidation step has been studied. Laccase-mediator couple has been shown as a reliable and mild system for the *N*-deprotection of *p*-methoxyphenyl (PMP) protected amines.[68] Thus, oxidation of the protected amine by the laccases from *Trametes versicolor* or *Agaricus bisporus* leads to formation of *p*-benzoquinone imine, which spontaneously hydrolyses in the aqueous reaction medium, furnishing the free amine and *p*-quinone, as shown in Scheme 18, using 0.92 mmol of the PMP-protected amine. Although high yields were obtained by employing the sole laccase, the use of catalytic amount of mediators could expand the scope of this methodology, demonstrated in the deprotection of *N*-PMP protected 4-phenylbutan-2-amine (**41**), a non-benzylic amine, where no conversion was observed without mediator. Best results were attained using violuric acid as a mediator after 48 h, achieving 88% conversion.



Scheme 18. Laccase-mediator (violuric acid) catalysed *N*-deprotection of *N*-PMP-protected amines.[68]

Another example of amine deprotection comprises a laccase-mediator system in a similar fashion, but in this case employing TEMPO as organic electron mediator.[69] In this work, *N*-benzylamines can be deprotected in high chemo- and regioselectivity. Thus, oxidation of the amine transiently affords the corresponding imine, which is spontaneously hydrolysed, delivering the free amine. Hence, using laccase of *Trametes versicolor* and TEMPO, reaction with *N*,*N*'-dibenzyl-4-amino-piperidine was highly regioselective through the secondary amine, obtaining as only product the corresponding free primary amine with complete conversion. Likewise, (±)-*trans*-*N*,*O*-dibenzyl-2-hydroxy-cyclohexylamine was successfully *N*-deprotected in a chemoselective fashion, yielding the *O*-benzyl derivative as sole product.

Taking advantage of laccases as mild and green oxidants, the synthesis of different disulfides **42** has been explored by homocoupling of heterocyclic thiols (Scheme 19).[70] Thus, employing catalytic ABTS as the organic mediator and the laccase from *Trametes versicolor*, one-electron oxidation leads to a thiyl radical, which collapses into the corresponding disulfide. Reactions were carried out in acetate buffer pH 4.4 containing MeOH (10% v v^{-1}). Disulfides were formed with yields from 50 to 95% depending on the substrate structure, without formation of overoxidation side products.



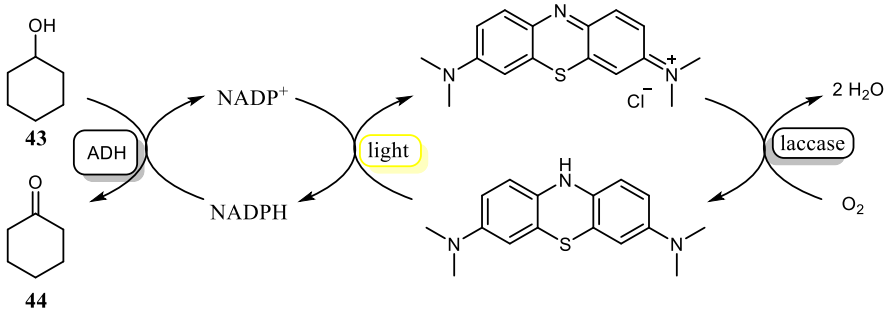
Scheme 19. Synthesis of a set of disulfides **42** employing laccase from *T. versicolor* in presence of ABTS as mediator.[70]

This kind of laccase/mediator systems have been also involved in alcohol oxidation. The oxidation of primary alcohols has been pursued in order to couple a secondary reaction employing *in-situ* the newly formed aldehyde.[71] A 1-4-diol or 1-5 diol can be oxidised by laccase/TEMPO, leading to a hydroxyaldehyde, that immediately cyclise affording a hemiacetal. Further oxidation by the same system allows the formation of stable butyro- and valerolactones in a one-pot one-step fashion.

4.3 One-pot catalytic combined redox processes driven by light.

In certain cases, chromophoric organic redox mediators (here considered organocatalysts) can be readily employed in biocatalysed processes under light irradiation. Photostimulation provides the chromophoric organocatalyst with a suitable redox potential to accept/donate electrons from/to molecules that otherwise shall be difficult or even impossible. In this way, flavin and analogues have been successfully used as excitable organocatalysts with fair turnover when coupled to oxidative enzymatic transformations.

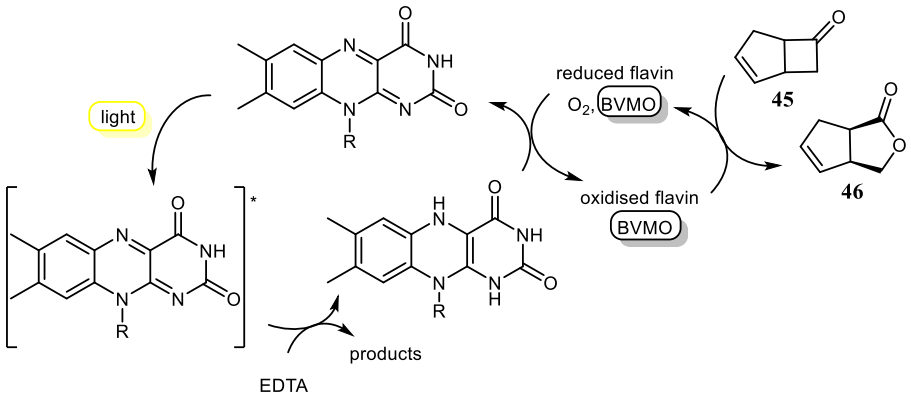
Likewise, it has been recently reported that pyrimidine cofactor regeneration to the oxidised form ($NAD(P)^+$) is feasible employing *Myceliophthora thermophila* laccase, (*Mtlac*), O_2 as final electron acceptor and dyes (Methylene blue, Methylene green and Azure B) as mediators, as shown in Scheme 20.[72] In this report, the authors accomplished to demonstrate that upon visible light irradiation, the NADH oxidation is three order of magnitude faster than the obscure counterpart. The authors coupled this $NAD(P)^+$ regeneration system with two enzymes, namely *Thermus* sp. ATN1 ADH, and a commercially available glucose dehydrogenase (GDH) to successfully obtain more than 30% conversion in two hours of cyclohexanone (**44**) from cyclohexanol (**43**) and gluconic acid from glucose, respectively. In the latter case, methylene blue was employed as organocatalyst showing a remarkable TTN of 2500.



Scheme 20. Use of a bienzymatic system laccase-ADH coupled with dyes as mediators for the photooxidation of cyclohexanol to cyclohexanone.[72]

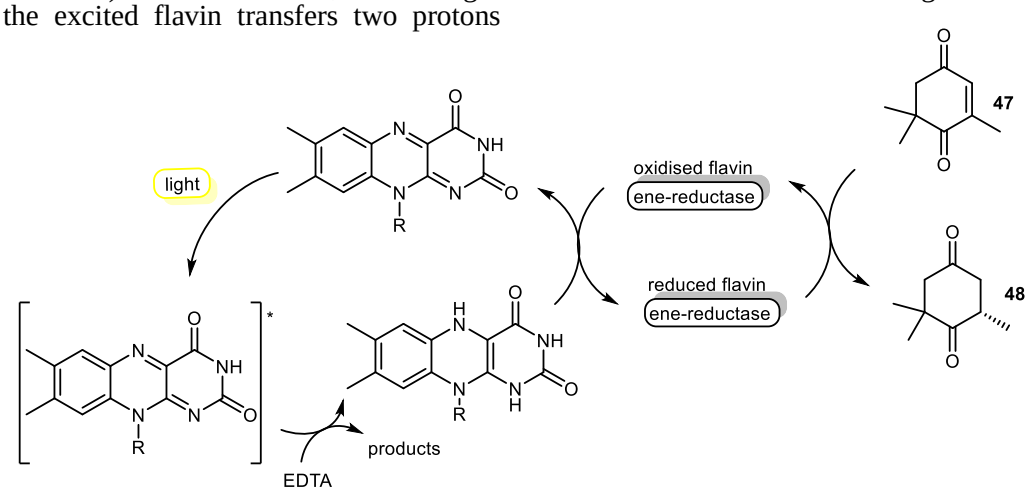
A similar approach has been showed in the *Caldariomyces fumago* chloroperoxidase (CPO)-catalysed sulfoxidation of thioanisole to enantiopure (*R*)-methyl phenyl sulfoxide employing FMN as chromophoric organocatalyst, EDTA as electron donor and molecular oxygen under photostimulation.[73] Hence, EDTA successively transfers electrons to the photoexcited FMN organocatalyst to be delivered to the heme prostetic group of CPO. The authors improve biocatalyst stability and mass transfer issues by adopting a biphasic surfactant-stabilised system which permits and increase in substrate concentration and sulfoxide productivity.

Similarly, EDTA/flavin in the presence of light has been employed in the preparation of lactones starting from cyclic ketones catalysed by BVMOs (Scheme 21).[74] In this way, the NADPH cofactor is replaced by the system EDTA/flavin, so, electrons coming from the sacrificial donor, pass to the diffusing excited flavin. Then, electrons are delivered to the BVMO-bound flavin to trigger formation of a enzyme-bound peroxyflavin, the actual oxo-transfer catalyst that finally introduces oxygen into the substrate. By using this methodology, a set of enantioenriched (>96 % *ee*) γ- and ε-lactones were successfully prepared from the corresponding prochiral ketones.



Scheme 21. EDTA/Flavin system employed in combination with BVMOs to catalyse Baeyer-Villiger oxidations. In the scheme, the oxidation of bicyclo[3.2.0]hept-2-en-6-one (**45**) to the corresponding lactone **46**.[74]

On the contrary, reductive enzymatic reactions combined with light-driven organocatalysed electron supply are even less explored than the already described oxidative counterparts. A representative example is the use EDTA/flavin in the presence of light for the reduction of electron deficient olefins into the corresponding saturated products by the action of an ene-reductase from *Bacillus subtilis* YqjM (Scheme 22).[75] In this case, under light irradiation the excited flavin transfers two protons and two electron from EDTA to the enzyme-bound flavin (prostetic group). Thus, YqjM with the prostetic group in its reduced form is able to deliver a hydride to the beta position of the C=C in a Michael-type addition reaction. In this report, ketoisophorone (**47**) was chosen as model substrate and the reduction to diketone **48** took place with a similar stereoselectivity than the standard reaction using NADPH and normal cofactor regeneration system.



Scheme 22. Reduction of ketoisophorone **47** catalysed by the ene-reductase from *Bacillus subtilis* YqjM in presence of EDTA and light.[75]

5 Outlook

Although organocatalysis and biocatalysis have reached a high degree of sophistication, as shown in the examples described in the present review. The interplay of both worlds, and therefore virtues from both sides, need further exploitation. The advancement in this area of catalysis shall necessarily come by adopting interdisciplinary research, in which enzyme engineering will provide biocatalysts with enhanced robustness towards organic solvents and reaction temperature, while molecular design will bring novel organocatalysts that may exert high asymmetric induction in a temperature range that matches that of the involved enzymes. In the field of organocatalysis, aside from the system lipase/carboxylic acid/peroxide, in most of the reported examples concerning combination with biocatalysis, the organocatalyst employed behaves as Lewis base.Therefore, it is tempting to explore novel cascade processes by targeting the use of Brønsted acid organocatalysts coupled with acidophilic enzymes. Moreover, metal-based catalyst can be efficiently incorporated into multicatalytic processes, thus allowing cross-coupling and cycloaddition reactions to take place concurrently with the organocatalytic and enzymatic transformations. The increasing number of artificial metalloenzymes,[76]

conceptually support this idea and in the near future may find broader partnerships in this area.[77] The employment of other technologies in combination with organo- and biocatalysis is worthy of further investigation. It can be envisioned more complex photochemical approaches,[78] that permit redox reactions to take place either on a catalyst or on a substrate/intermediate with great deal of success. Finally, a general and reliable asymmetric version of lipase/carboxylic acid/peroxide system is still elusive and a careful design of the carboxylic acid organocatalyst will be required.[79]

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References

[1] a) E. García-Junceda, I. Lavandera, D. Rother, J. H. Schrittwieser, *J. Mol. Catal. B: Enzym*. **2015**, *114*, 1-6. b) S. F. Mayer, W. Kroutil, K. Faber, *Chem. Soc. Rev.*, **2001**, *30*, 332-339; c) F. R. Bisogno, I. Lavandera, V. Gotor, Biocatalytic Concurrent Processes. *Kirk-Othmer Encyclopedia of Chemical Technology*, John Wiley & Sons, Hoboken, **2011**, 1–20; d) D. L. Nelson, M. Cox, *Lehninger: Principles of Biochemistry*, W. H. Freeman, New York, **2004**; e) H. Curtis, N. S. Barnes, *Biology*, W. H. Freeman, New York, **1989**.

[2] a) R. A. Sheldon, *Pure Appl. Chem*. **2000**, *72*, 1233-1246; b) B. M. Trost, *Science* **1991**, *254*, 1471-1477.

[3] See: a) F. R. Bisogno, A. Rioz-Martínez, C. Rodríguez, I. Lavandera, G. de Gonzalo, D. E. Torres-Pazmiño, M. W. Fraaije, V. Gotor, *ChemCatChem* **2010**, *2*, 946-949; b) C. V. Voss, C. C. Gruber, W. Kroutil, *Angew. Chem*. **2008**, *120*, 753-757; *Angew. Chem. Int. Ed*. **2008**, *47*, 741-745; c) J.-C. Wasilke, S. J. Obrey, R. T. Baker, G. C. Bazan, *Chem. Rev*. **2005**, *105*, 1001-1020.

[4] F. G. Mutti, A. Orthaber, J. H. Schrittwieser, J. G. de Vries, R. Pietschnig, W. Kroutil, *Chem. Commun*. **2010**, *46*, 8046-8048.

[5] a) A. Cuetos, F. R. Bisogno, I. Lavandera, V. Gotor *Chem. Commun*. **2013**, *49*, 2625-2627; b) G. de Gonzalo, G. Ottolina, G. Carrea, M. W. Fraaije, *Chem. Commun*. **2005**, 3724-3726.

[6] See for instance: a) S. Rossi, M. Benaglia, E. Massolo, L. Raimondi, *Catal. Sci. Technol*. **2014**, *4*, 2708-2723; b) P. I. Dalko in *Comprenhensive Enantioselective Organocatalysis: Catalysts, Reactions and Applications*, Wiley-VCH, Weinheim, **2013**; c) B. List (Ed.), Asymmetric Organocatalysis, Springer Verlag, Berlin Heidelberg, **2009**; d) A. Dondoni, A. Massi, *Angew. Chem*. **2008**, *120*, 4716-4739; *Angew. Chem. Int. Ed*. **2008**, *47*, 4638-4660; e) A. Berkessel, H. Gröger in *Asymmetric Organocatalysis: From Biomimetic Concepts to Applications in Asymmetric Synthesis*, Wiley-VCH, Weinheim, **2005**; f) P. I. Dalko, L. Moisan, *Angew. Chem*. **2004**, *116*, 5248-5286; *Angew. Chem. Int. Ed*. **2004**, *43*, 5138–5175;

[7]a) R. N. Patel in *Green Biocatalysis*, John Wiley& Sons, Hoboken, **2016**; b) C. N. Clouthier, J. N. Pelletier, *Chem. Soc. Rev*.**2012**, *41*, 1585-1605; c)U. T. Bornscheuer, G. W. Huisman, R. J. Kazlauskas, S. Lutz, J. C. Moore, K. Robins, *Nature***2012**, *485*, 185-194; d) K. Faber in *Biotransformations in Organic Chemistry*, Springer, Berlin-Heidelberg, 6th Edition, **2011**; e) G. A. Behrens, A. Hummel, S. K. Padhi, S. Schätzle, U. W. Bornscheuer, *Adv. Synth. Catal*. **2011**, *353*, 2191-2215.

[8] For excellent surveys and discussions, see: a) C. A. Denard, J. F. Hartwig, H. Zhao, *ACS Catal*. **2013**, *3*, 2856-2864;b) B. Martín-Matute, J.-E. Bäckvall, *Curr. Opin. Chem. Biol*.**2007**, *11*, 226-232;c) Y. Ahn, S.-B. Ko, M.-J. Kim, J. Park, *Coord. Chem. Rev*. **2008**, *252*, 647-658; d) O. Pamies, J.-E. Bäckvall, *Chem. Rev*. **2003**, *103*, 3247-3262.

[9] F. Zhou, Y-L. Liu, J. Zhou in *Multicatalyst System in Asymmetric Catalysis, 1st ed*., (Ed.: J. Zhou), John Wiley& Sons, Hoboken,**2015**, pp 501-631, and references therein.

[10] For an outstanding evaluation of the state of the art, see:H. Gröger,W. Hummel, *Curr. Opin. Chem. Biol*. **2014**, *19*, 171-179.

[11]Some examples of additives and solvent effects on organo- and biocatalysed reactions: a) A. Martínez-Castañeda, K. Kezdora, I. Lavandera, H. Rodríguez-Solla, C. Concellón, V. del Amo, *Chem. Commun*. **2014**, *50*, 5298-5260; b) C. M. Clouthier, J. N. Pelletier, *Chem. Soc. Rev*. **2012**, *41*, 1585-1605; c) N. El-Hamdouni, X. Companyó, R. Ríos, A. Moyano, *Chem. Eur. J*. **2010**, *16*, 1142-1148; d) C. Rodríguez, G. de Gonzalo, M. W. Fraaije, V. Gotor, *Green. Chem*. **2010**, *12*, 2255-2260; e) G. de Gonzalo, G. Ottolina, F. Zambianchi, M. W. Fraaije, G. Carrea, *J. Mol. Catal. B: Enzym*. **2006**, *39*, 91-97; f) S. Riva, G. Carrea, *Angew. Chem*. **2000**, *112*, 2312-2341; *Angew. Chem. Int. Ed*. **2000**, *39*, 2226-2254.

[12] See for example: E. García-Junceda in *Multi-Step Enzyme Catalysis: Biotransformations and Chemoenzymatic Synthesis, 1st ed*., Wiley-VCH, Weinheim, **2008**.

[13] M. Edin, J.-E. Bäckvall, A. Córdova, *Tetrahedron: Lett.* 2004, 45, 7697-7701.

[14] For some examples on ADHs applications, see: a) C. M. Nealon, M. M. Musa, J. M. Patel, R. S. Phillips, *ACS Catal.***2015**, *5*, 2100-2114; b) M. M. Musa, R. S. Phillips, *Catal. Sci. Technol*. **2011**, *1*, 1311-1323.

[15] M. Heidlindemann, M. Hammel. U. Scheffler, R. Mahrwald, W. Hummel, A. Berkessel, H. Gröger, *J. Org. Chem*. **2015**, *80*, 3387-3396.

[16] C.-S. Da, L.-P.Che, Q.-P.Guo, F.-C.Wu, X. Ma, Y.-N. J. Jia, *J.Org. Chem*. **2009**, *74*, 2541−2546.

[17] a) R. Wohlgemuth, *New Biotechnol*. **2009**, *25*, 204-213; b) E. M. Buque-Taboada, A. J. J. Straathoft, J. J. Heijinen, L. A. M. van der Wielen, *Appl. Microbiol. Biotechnol.***2006**, *71*, 1-12; c) G. J. Lye, J. M. Woodley, *Trends Biotechnol*. **1999**, *17*, 395-402; d) A. Freeman, J. M. Woodley, M. D. Lilly, *Nature Biotechnol*. **1993**, *11*, 1007-1012.

[18] K. Baer, M. Krauβer, E. Burda, W. Hummel, A. Berkessel, H. Gröger, *Angew. Chem*. **2009**, *121*, 9519-9522; *Angew. Chem. Int. Ed*. **2009**, *48*, 9355-9338.

[19] M. Raj, Vishnumaya, S. K. Ginotra, V. K. Singh, *Org. Lett.* **2006**, *8*, 4097-4099.

[20] G. Rulli, N. Dunangdee, K. Baer, W. Hummel, A. Berkessel, H. Gröger, *Angew. Chem*. **2011**, *123*, 8092-8095; *Angew. Chem. Int. Ed.* **2011**, *50*, 7944-7947.

[21] M. Heidlindemann, G. Rulli, A. Berkessel. W. Hummel, H. Gröger, *ACS Catal*. **2014**, *4*, 1099-1103.

[22] See for instance: a) T. E. Kristensen, T. Hansen, *Eur. J. Org. Chem.***2010**, 3179-3204; b) F. Cozzi, *Adv. Synth. Catal*. **2006**, *348*, 1367-1390.

[23] G. E. Jeromin, *Biotechnol. Lett*. **2009**, *31*, 1717-1721.

[24] R. C. Simon, E. Busto, J. H. Schrittwiesser, J. H. Sattler, J. Pietruszka, K. Faber, W. Kroutil, *Chem. Commun*. **2014**, *50*, 15669-15672.

[25] a) K. Akagawa, K. Kudo, *Adv. Synth. Catal.* **2011**, *353*, 843-847; b) K. Akagawa, T. Fujiwara, S. Sakamoto, K. Kudo, *Chem. Commun*. **2010**, *46*, 8040-8042.

[26] K. Akagawa, R. Umezawa, K. Kudo, *Beilstein J. Org. Chem*. **2012**, *8*, 1333-1337.

[27] For some reviews on laccases, see: a) U. P. Dwivedi, P. Singh, V. P. Pandey, A. Kumar, *J. Mol. Catal. B: Enzym*. **2011**, *68*, 117-128; b) F. Hollmann, I. W. C. E. Arends, K. Buehler, *ChemCatChem* **2010**, *2*, 762-782; c) S. Witayakran, A. J. Ragauskas, A. J. *Adv. Synth. Catal*. **2009**, *351*, 1187-1209; d) S. Riva, *Trends Biotechnol*. **2006**, *24*, 219-226.

[28] A. Kinnell, T. Harman, M. Bingham, A. Berry, A. Nelson, *Tetrahedron* **2012**, *68*, 7719-7722.

[29] T. Katnemitsu, M. Inoue, N. Yoshimura, K. Yoneyama, R. Watarai, M. Miyazaki, Y. Odanaka, K. Nagata, T. Itoh, *Eur. J. Org. Chem*. **2016**, 1874-1880.

[30] See for example: P.E. Schreiner, A. Wittkopp, *Org. Lett*. **2002**, *4*, 217-220.

[31] G. Rulli, N. Duangdee, W. Hummel, A. Berkessel, H. Gröger, *Eur. J. Org. Chem*. **2017**, 812-817.

[32] C. R. Müller, I. Meiners, P. Domínguez de María, *RSC Adv.* **2014**, *4*, 46097-46101.

[33] For reviews on DES and their application in catalysis, see for example: a) N. Guajardo, C. R. Müller, R. Schrebler, C. Carlesi, P. Domínguez de María, *ChemCatChem*, **2016**, *8*, 1020-1027; b) M. Krystof, M. Pérez-Sánchez, P. Domínguez de María, *ChemSusChem*, **2013**, *6*, 630-634;c) D. Carriazo, M. C. Serrano, M. C. Gutiérrez, M. L. Ferrer, F. del Monte, *Chem. Soc. Rev*. **2012**, *41*, 4996-5014.

[34] G. A. Hafenstine, K. Ma, A. W. Harris, O. Yehezkeli, E. Park, D. W. Domaille, J. N. Cha, A. P. Goodwin, *ACS Catal.* **2017**, *7*, 568-572.

[35]K. Akagawa, K. Kudo, *Org. Lett.* **2011**, *13*, 3498-3510.

[36] P. S. Baran, M. P. DeMartino, *Angew. Chem. Int. Ed*. **2006**, *45*, 7083-7086; *Angew. Chem*. **2006**, *118*, 7241-7244; b) S. P. Brown, M. P. Brochu, C. J. Sinz, D. W. C. MacMillan, *J. Am. Chem. Soc*. **2003**, *125*, 10808-10809.

[37] S. Sujić, J. Pietruszka, D. Worgull, *Adv. Synth. Catal*. **2015**, *357*, 1822-1830.

[38] J. P. M. Nunes, C. A. M. Afonso, S. Caddick, *Tetrahedron: Lett*. **2009**, *50*, 3706-3708.

[39] Some reviews on biocatalytic and organocatalytic DKRs: a) P. Li, X. Hu, X.-Q. Dong, X. Zhang, *Molecules***2016**, *10*, 1327; b) O. Langvik, T. Saloranta, D. Y. Murzin, R. Leino, *ChemCatChem***2015**, *7*, 4004-4015; c) P. Hoyos, V. Pace, V. Alcántara, *Adv. Synth. Catal.***2012**, *354*, 2585-2611; d) H. Pellisier, *Adv. Synth.Catal*.**2011**, *353*, 659-676; e) J. H. Lee, K. Han, M. J. Kim, J. Park, *Eur. J. Org. Chem*. **2010**, 999-1015.

[40] J. P. M. Nunes, L. F. Veiros, P. D. Vaz, C. A. M. Afonso, S. Caddick, *Tetrahedron***2011**, *67*, 279-2787.

[41] M. Anderson, S. Afewerki, P. Berglund, A. Córdova, *Adv. Synth. Catal*. **2014**, *356*, 2113-2118.

[42] M. Ikagi, J. Hiratake, T. Nishioka, J. Oda, *J. Am. Chem. Soc.* **1991**, *113*, 9360-9361.

[43] C. Paizs, P. Tähtinen, K. Lundell, L. Poppe, F.-D, Irimie, L. T. Kanerva, *Tetrahedron: Asymmetry***2003**, *14*, 1895-1904.

[44] C. Paizs, P. Tähtinen, K. Lundell, L. Poppe, F.-D, Irimie, L. T. Kanerva, *Tetrahedron: Asymmetry***2003**, *14*, 619-627.

[45] Y.-X. Li, A. J. J. Straathof, U. Hanefeld, *Tetrahedron: Asymmetry***2002**, *13*, 739-743

[46] L. Veum, U. Hanefeld, *Tetrahedron: Asymmetry***2004**, *15*, 3707-3709.

[47] P. Vongvilai, R. Larsson, O. Ramström, *Adv. Synth. Catal.***2008**, *350*, 448-452.

[48] For some reviews on BVMOs see: a) M. Bucko, P. Gemeiner, A. Schenkmayerova, T. Krajcovic, F. Rudroff, M. D. Mihovilovic, *Appl. Microbiol. Biotechnol.* **2016**, *100*, 6585-6599; b) G. de Gonzalo, W. J. H. van Berkel, M. W. Fraaije in *Science of Synthesis, Biocatalysis Vol. 3*, (Eds.: K. Faber, N. J. Turner, W. D. Fessner), Georg Thieme-Verlag, Suttgart, **2015**, pp 187-234; c) H. Leisch, K. Morley, P. C. K. Lau, *Chem. Rev.* **2011**, *111*, 4165-4222;

[49] N.M. Kamerbeek, A. J. J. Olsthoorn, M.W. Fraaije, D. B. Janssen,*Appl. Environ. Microbiol*.**2003**, *69*, 419-426.

[50] C. Rodríguez, G. de Gonzalo, A. Rioz-Martínez, D. E. Torres Pazmiño, M. W. Fraaije, V. Gotor, *Org. Biomol. Chem.* **2010**, *8*, 1121-1125.

[51] D. E. Torres Pazmiño R. Snajdrova, D. V.Rial, M. D. Mihovilovic, M. W. Fraaije, *Adv. Synth. Catal*. **2007**, *349*, 1361-1368.

[52] G. de Gonzalo, C. Rodríguez, A. Rioz-Martínez, V. Gotor, *Enzyme Microb. Technol*. **2012**, *50*, 43-49.

[53] See for instance: G. A. Applegate, D. B. Berkowitz, *Adv. Synth. Catal*. **2015**, *357*, 1619-1632.

[54] J. Mangas-Sánchez, E. Busto, V. Gotor, V. Gotor-Fernández, *Org. Lett*. **2013**, *15*, 3872-3875.

[55] D. Méndez-Sánchez, J. Mangas-Sánchez, E. Busto, V. Gotor, V. Gotor-Fernández, *Adv. Synth. Catal*. **2016**, *358*, 122-131.

[56] P. Bongen, J. Pietruszka, R. C. Simon, *Chem. Eur. J*. **2012**, *18*, 11063-11070.

[57] T. Ishikawa in *Superbases for Organic Synthesis*, Wiley-VCH, Weinheim, **2009**.

[58] For selected examples, see: a) K. Zalewska, M. M. Santos, H. Cruz, L. C. in *Recent Advances in Organocatalysis*(Eds.: I Karamé, H. Srour), InTech, **2016**; b) M. C. A.van Vliet, I. W. C. E.Arends, R.A.Sheldon, *Chem. Commun.* **1999**, 263-264;c) M. Frohn, X. Zhou, J.-R. Zhang,Y. Tang, Y.Shi, *J. Am. Chem. Soc.***1999**, *121*, 7718-7719;d) V. K. Aggarwal, M. F. Wang, *Chem. Commun*. **1996**, 191-192.

[59] For selected examples, see: a) A. J. Carnell, W. Clegg, R. A. W. Johnstone, C. C. Parsy, W. R. Sanderson, *Tetrahedron* **2000**, *56*, 6571-6575; b) P. Lupattelli, R. Ruzziconi, P. Scafato, A. Degl'Innocenti, A. B. Paolobelli, *Synth. Commun*. **1997**, *27*, 441-446; c) G. Hanquet, X. Lusinchi, *Tetrahedron Lett*. **1993**, *34*, 5299-5302.

[60] C.Palomo, J. M. Aizpurua, C. Cuevas,R. Urchegui, A. Linden, *J. Org. Chem*. **1996**, *61*, 4400-4404.

[61] J.-W. Yang, M. T. Hechavarría Fonseca, N. Vignola, B. List,*Angew. Chem*. **2005**, *117*, 110-112; *Angew. Chem. Int. Ed*. **2005**, *44*, 108–110.

[62] For comprehensive reviews, see a) D. Holtmann, M. W. Fraaije, I. W. C. E. Arends, D. J. Oppermand F. Hollmann, *Chem. Commun*. **2014**, *50*, 13180-13200; b) G. de Gonzalo, A. A. Orden, F. R. Bisogno, *Curr. Org. Chem*. **2012**, *16*, 2598-2612; c) F. Hollmann, I. W. C. E. Arends, K. Buehler, A. Schallmey, B. Bühler, *Green Chem*. **2011**, *13*, 226-265; e) R. D. Schmid, V. B. Urlacher (Eds.), *Modern Biooxidation: Enzymes, Reactions and Applications*, Wiley-VCH, Weinheim, **2007**.

[63] F. Björkling, S. E. Godtfredsen, O. Kirk, *J. Chem. Soc. Chem. Commun.* **1990**, 1301-1303.

[64] For didactical surveys, see: a) M. López-Iglesias, V. Gotor-Fernández, *Chem. Rec*. **2015**, *15*, 743-759; b)E. Busto, V. Gotor-Fernández, V. Gotor, *Chem. Soc. Rev.* **2010**, *39*, 4504-4523; c) U. T. Bornscheuer, R. J. Kazlauskas, *Angew. Chem*. **2004**, *116*, 6156-6165; *Angew. Chem. Int. Ed.* **2004**, *43*, 6032-6040.

[65] R. Madeira Lau, F. van Rantwijk, K. R. Seddon, R. A. Sheldon, *Org. Lett*. **2000**, *2*, 4189-4191.

[66] For selected examples of catalytic version of lipase/carboxylic acid-mediated Baeyer-Villiger oxidation and sulfoxidation, respectively, see: a) S. C. Lemoult, P. F. Richardson, S. M. Roberts, *J. Chem. Soc., Perkin Trans. 1*, **1995**, 89-91; b) F. Björkling, H. Frykman, S. E. Godtfredsen, O. Kirk, *Tetrahedron* **1992**, *48*, 4581-4592.

[67] W. Adam, C. R. Saha-Möller, P. A. Ganeshpure,*Chem. Rev.* **2001**, *101*, 3499-3548.

[68] J. M. M. Verkade, Lieke J. C. van Hemert, P. J. L. M. Quaedflieg, H. E. Schoemaker, M. Schürmann, F. L. van Delft, F. P. J. T. Rutjes. *Adv. Synth. Catal*. **2007**, *349*, 1332-1336.

[69] L. Martínez-Montero, A. Díaz-Rodríguez, V. Gotor, V. Gotor-Fernández, I. Lavandera. *Green. Chem*. **2015**, *17*,2794-2798.

[70] H. T. Abdel-Mohsen, K. Sudheendran, J. Conrad, U. Beifuss. *Green Chem*. **2013**, 15, 1490-1495.

[71] A. Díaz-Rodríguez, L. Martínez-Montero, I. Lavandera, V. Gotor, V. Gotor-Fernández, *Adv. Synth. Catal*. **2014**, *356*, 2321 - 2329.

[72] S. Kochius, Y. Ni, S. Kara, S. Gargiulo, J. Schrader, D. Holtmann, F. Hollmann, *ChemPlusChem* **2014**, *79*, 1554–1557.

[73] E. Churakova, I. W. C. E. Arends, F. Hollmann, *ChemCatChem* **2013**, *5*, 565-568.

[74] F. Hollmann, A. Taglieber, F. Schulz, M. T. Reetz, *Angew. Chem.***2007**, 119, 2961-2964; *Angew. Chem. Int. Ed*. **2007**, *46*, 2903-2906.

[75] M. M. Grau, J. C. van der Toorn, L. G. Otten, P. Macheroux, A. Taglieber, F. E. Zilly, I. W. C. E. Arends, F. Hollmann, *Adv. Synth. Catal*. **2009**, *351*, 3279–3286.

[76] For representative examples and discussions, see: a) M. Jeschek, R. Reuter, T. Heinisch, C. Trindler, J. Klehr, S. Panke, T. R. Ward, *Nature* **2016**,*537*, 661-665;b) I. Drienovská, A. Rioz-Martínez, A. Draksharapu, G. Roelfes, *Chem. Sci*. **2015**, *6*, 770-776; c) A. Rioz-Martínez, G. Roelfes, *Curr. Opin. Chem. Biol*., **2015**, *25*, 80-87; d) H. M. Key, P. Dydio, D. S. Clark, J. J. P. Peyralans, A. Maichele, Y. Fu, M. Maywald, *Chem. Commun*. **2006**, 4318-4320.

[77] V. Köhler, Y. M. Wilson,M. Dürrenberger, D. Ghislieri, E. Churakova, T. Quinto, L. Knörr, D. Häussinger, F. Hollmann ,N. J. Turner, T. R. Ward, *Nature Chem*. **2013**, *5*, 93-99.

[78] For outstanding reviews, see: a) N. A. Romero, D. A. Nicewicz, *Chem. Rev*. **2016**, *116*, 10075−10166; b) D. Ravelli, M. Fagnoni, A. Albini, *Chem. Soc. Rev*. **2013**, *42*, 97-113.

[79] Recently, an asymmetric lipase-catalysed perhydrolysis enabling asymmetric Baeyer Villiger oxidative desymmetrization of prochiral ketones was reported. In spite of the high conversion and remarkable enantioselectivity obtained, the carboxylic acid is not employed in substoichiometric amounts; instead, 200-400 % mol of this 'organocatalyst' was needed to achieve such results after 8 days of reaction. See: A. Drożdż, A. Chrobok, *Chem. Commun.* **2016**, *52*, 1230-1233.

REVIEW

Organocatalysis and biocatalysis hand in hand: combining catalysts in one-pot procedures

*Adv. Synth. Catal.***Year**, *Volume*, Page – Page

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